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PARENTAL RNAI SUPPRESSION OF CHROMATIN REMODELING GENES TO CONTROL COLEOPTERAN PESTS

Blair D. Siegfried
Lincoln, NE, bsiegfried1@unl.edu

Kenneth E. Narva
Zionsville, IN

Kanika Arora
Indianapolis, IN

Sarah E. Worden
Indianapolis, IN

Chitvan Khajuria
Chesterfield, MO

See next page for additional authors

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Authors

Blair D. Siegfried, Kenneth E. Narva, Kanika Arora, Sarah E. Worden, Chitvan Khajuria, Elane Fishilevich, Nicholas P. Storer, Meghan Frey, Ronda L. Hamm, and Ana Maria Velez



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(54) **PARENTAL RNAI SUPPRESSION OF
 CHROMATIN REMODELING GENES TO
 CONTROL COLEOPTERAN PESTS**

USPC 800/279
 See application file for complete search history.

(71) Applicants: **Dow AgroSciences LLC**, Zionsville, IN
 (US); **The Board of Regents of the
 University of Nebraska**, Lincoln, NE
 (US)

(72) Inventors: **Blair D. Siegfried**, Lincoln, NE (US);
Kenneth E. Narva, Zionsville, IN
 (US); **Kanika Arora**, Indianapolis, IN
 (US); **Sarah E. Worden**, Indianapolis,
 IN (US); **Chitvan Khajuria**,
 Chesterfield, MO (US); **Elane
 Fishilevich**, Indianapolis, IN (US);
Nicholas P. Storer, Kensington, MD
 (US); **Meghan Frey**, Greenwood, IN
 (US); **Ronda L. Hamm**, Carmel, IN
 (US); **Ana Velez**, Lincoln, NE (US)

(73) Assignees: **Dow AgroSciences LLC**, Indianapolis,
 IN (US); **The Board of Regents of the
 University of Nebraska**, Lincoln, NE
 (US)

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 16, 2014.

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C12N 15/113 (2010.01)
A01N 57/16 (2006.01)
A01N 63/02 (2006.01)

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 CPC **C12N 15/8286** (2013.01); **A01N 57/16**
 (2013.01); **A01N 63/02** (2013.01); **C12N**
15/113 (2013.01); **C12N 15/8218** (2013.01);
C12N 2310/14 (2013.01)

(58) **Field of Classification Search**
 CPC **C12N 15/8286**

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Primary Examiner — Li Zheng

(74) *Attorney, Agent, or Firm* — Sean M. Russell;
 Magleby Cataxinos & Greenwood

(57) **ABSTRACT**

This disclosure concerns nucleic acid molecules and meth-
 ods of use thereof for control of hemipteran pests through
 RNA interference-mediated inhibition of target coding and
 transcribed non-coding sequences in hemipteran pests. The
 disclosure also concerns methods for making transgenic
 plants that express nucleic acid molecules useful for the
 control of hemipteran pests, and the plant cells and plants
 obtained thereby.

31 Claims, 11 Drawing Sheets

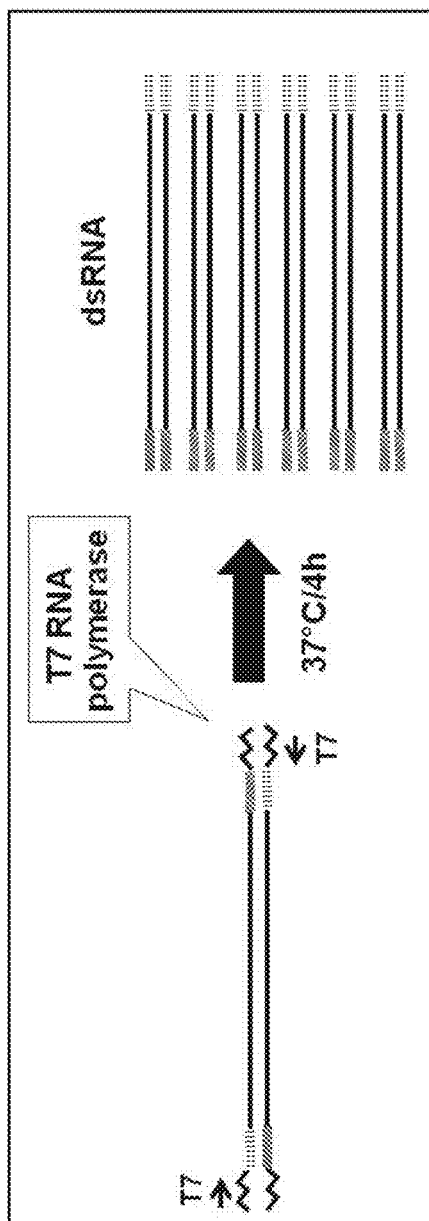


FIG. 1A

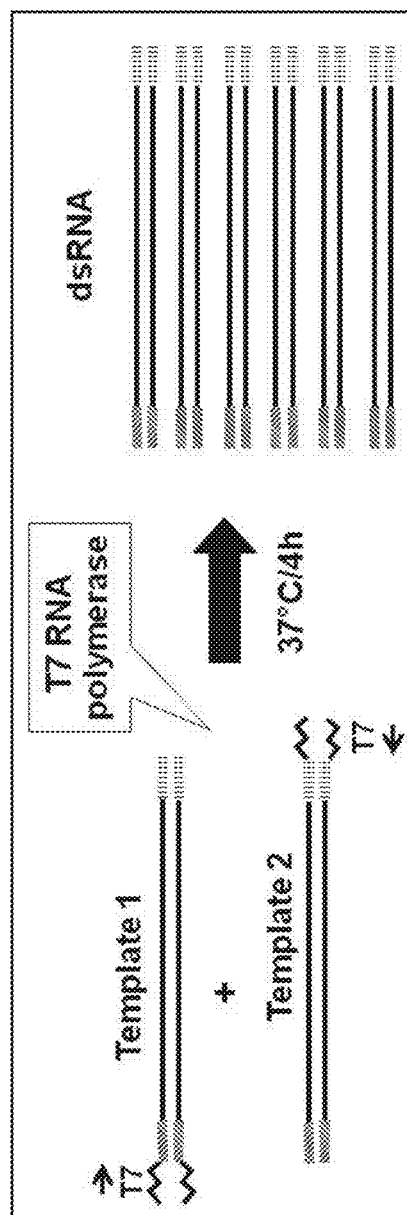
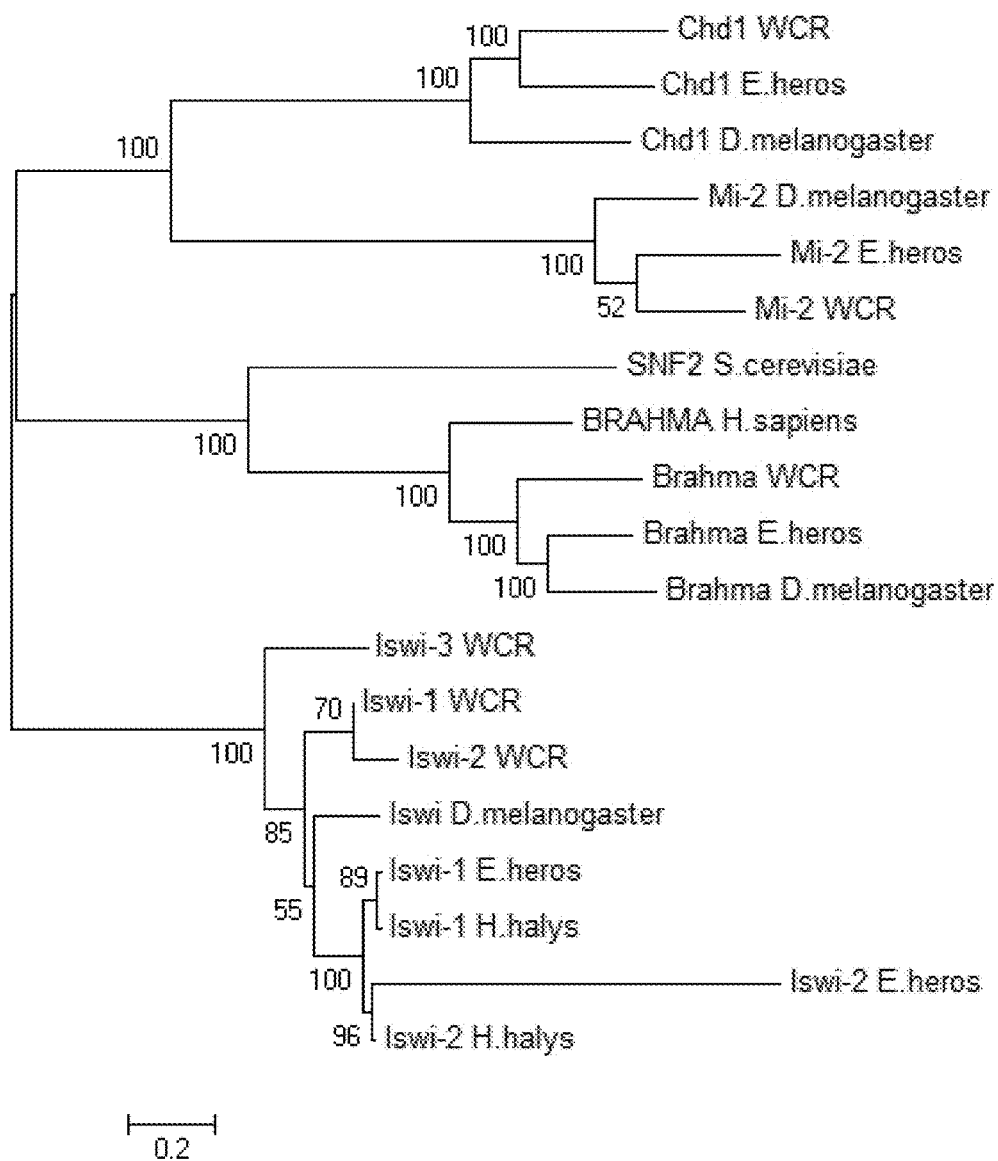


FIG. 1B

**FIG. 2**

SWI2/NSF2 family: brahma

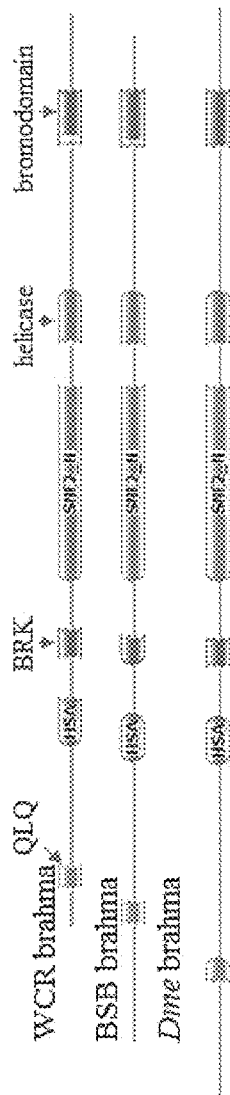


FIG. 3A

ISWI family: Iswi

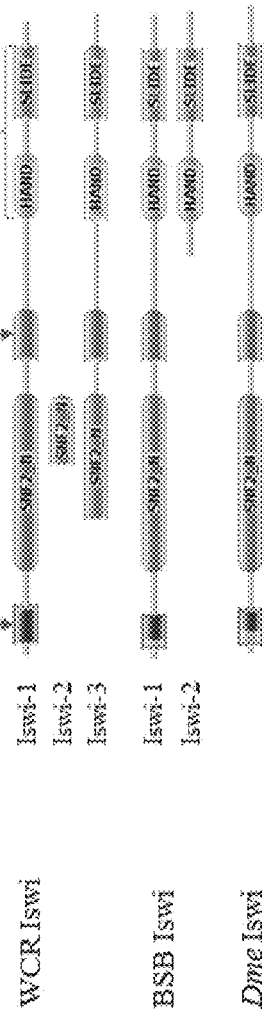


FIG. 3B

CHD subfamily I: Chd1

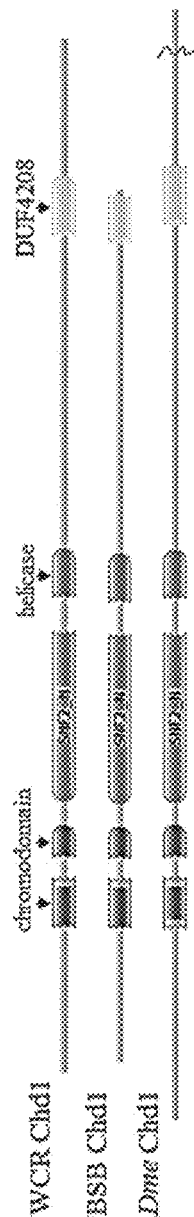


FIG. 3C

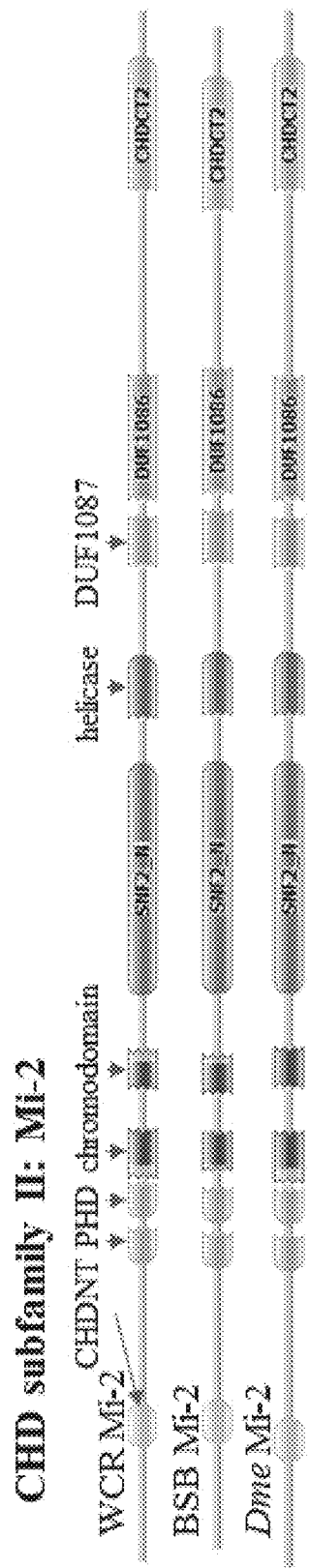


FIG. 3D

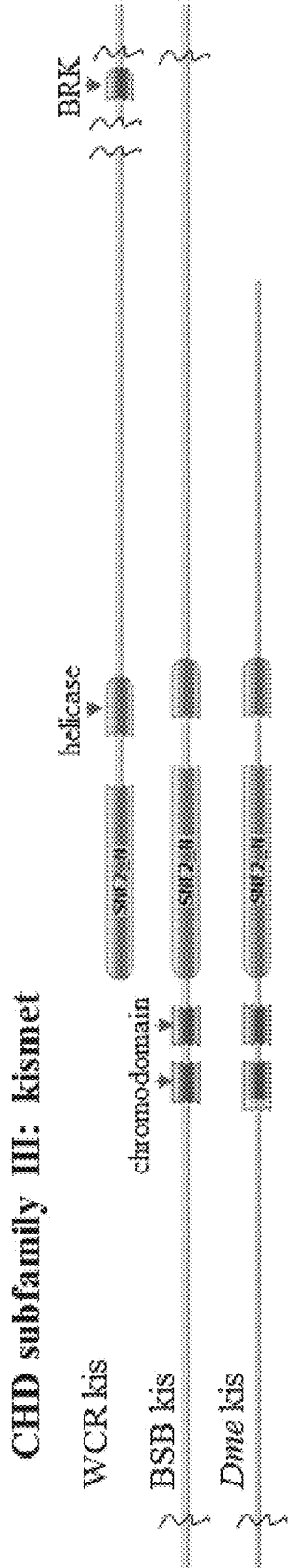


FIG. 3E

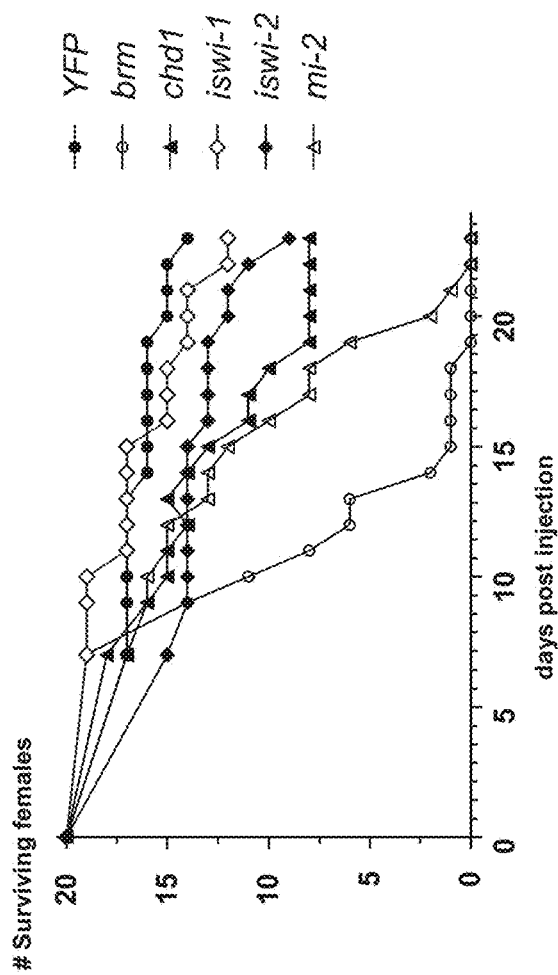


FIG. 4A

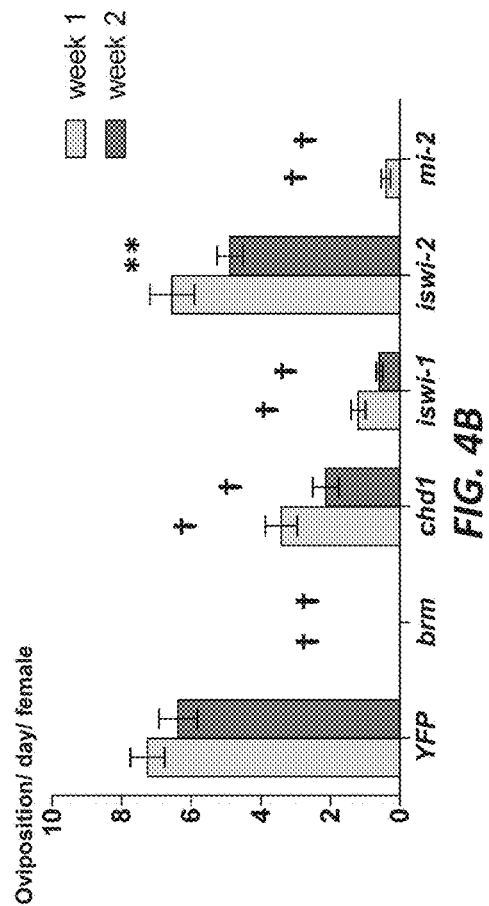


FIG. 4B

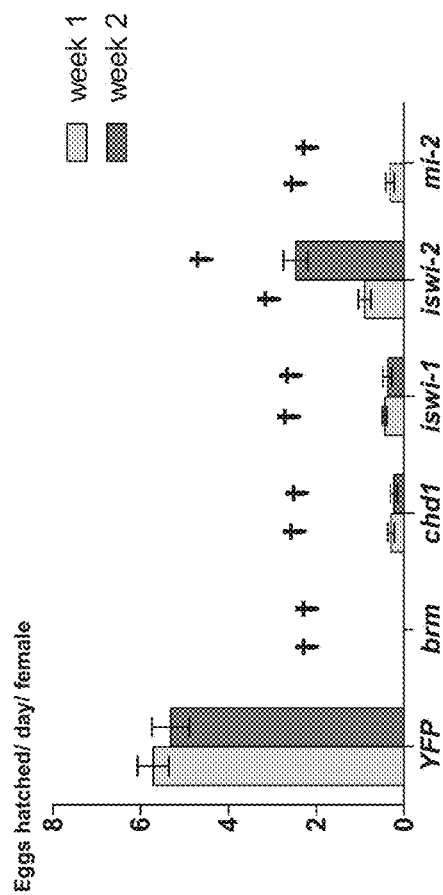


FIG. 4C

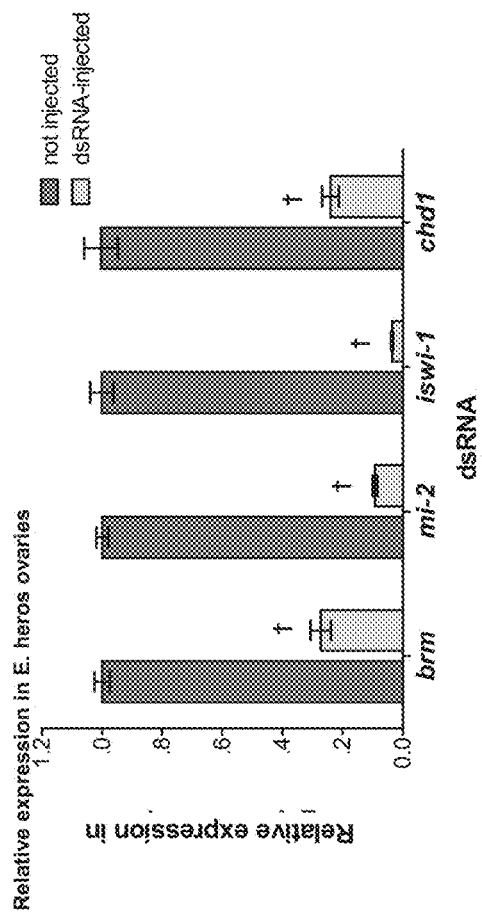
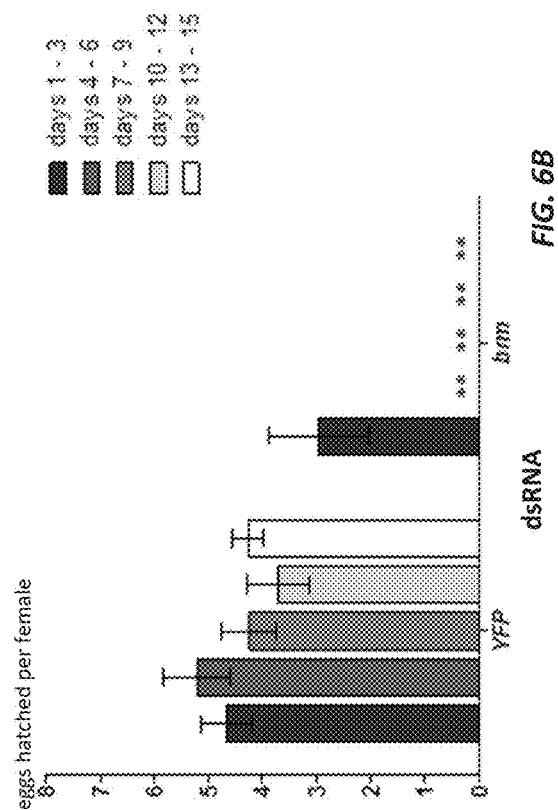
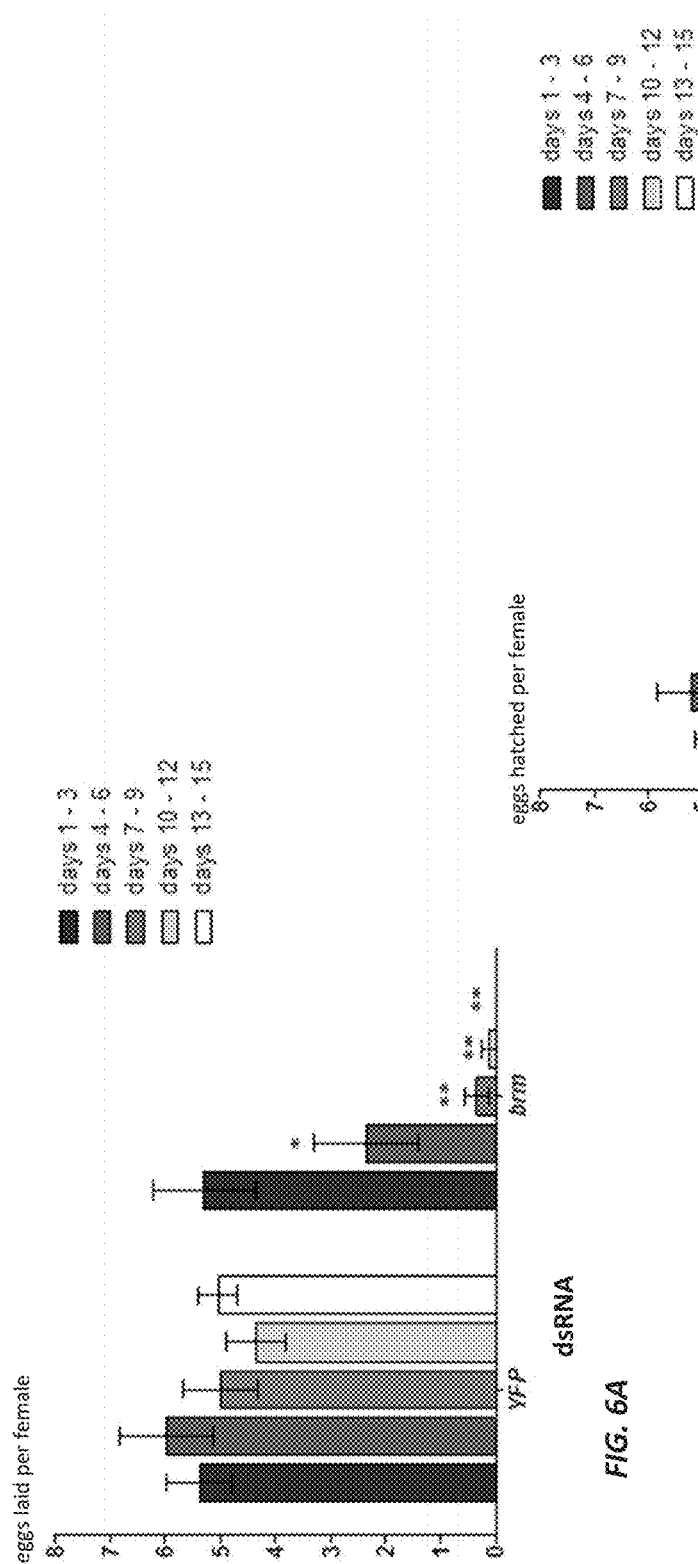
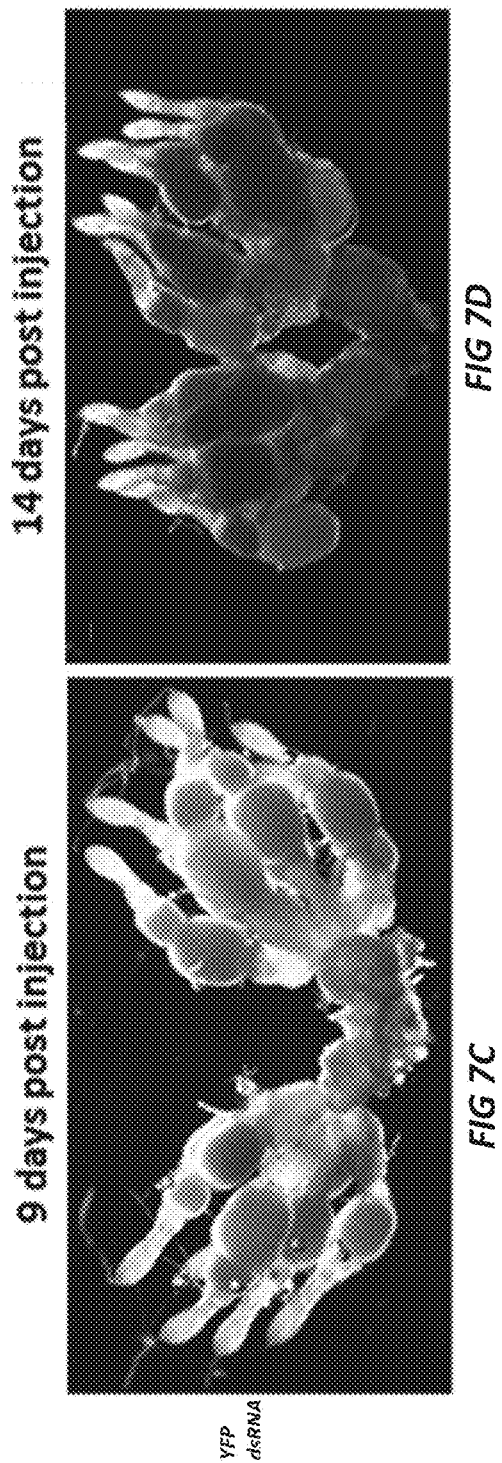
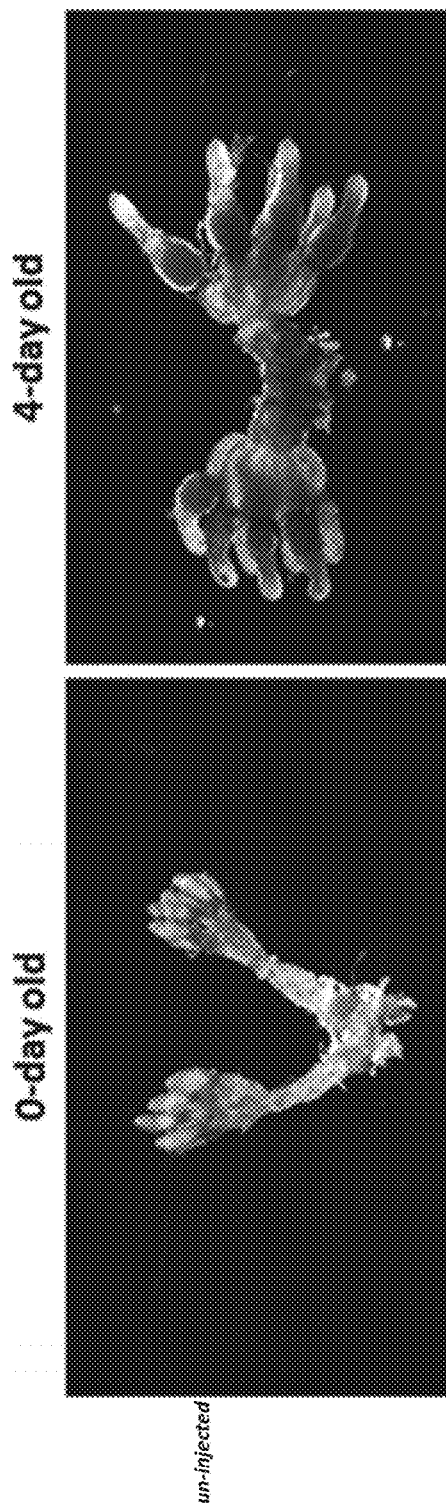


FIG. 5





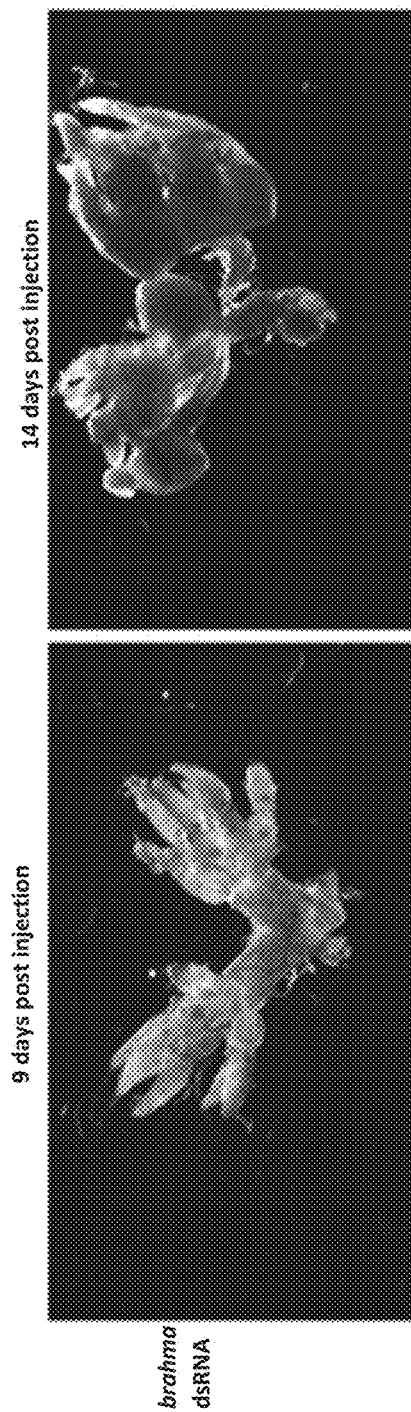


FIG 7E

FIG 7F

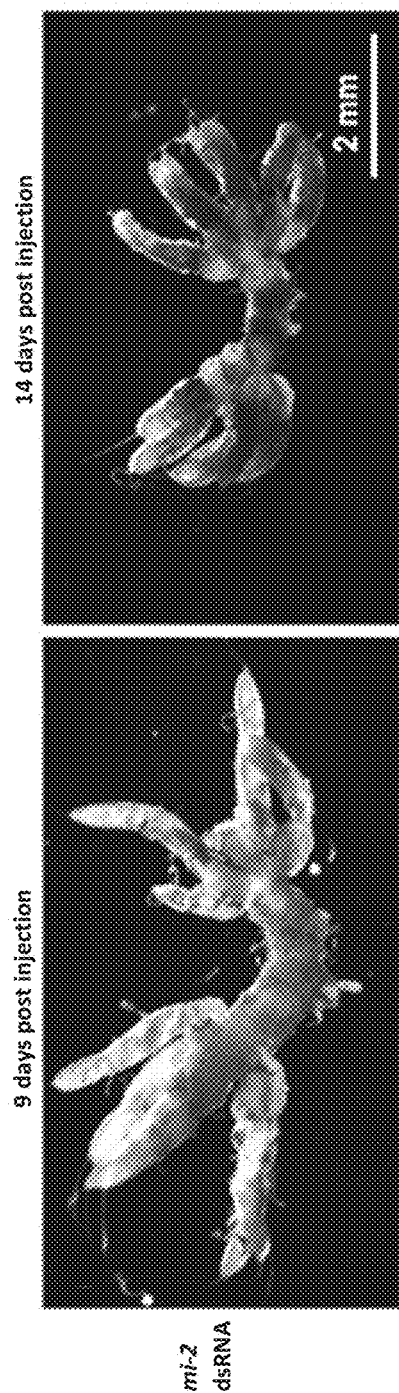


FIG 7G

FIG 7H

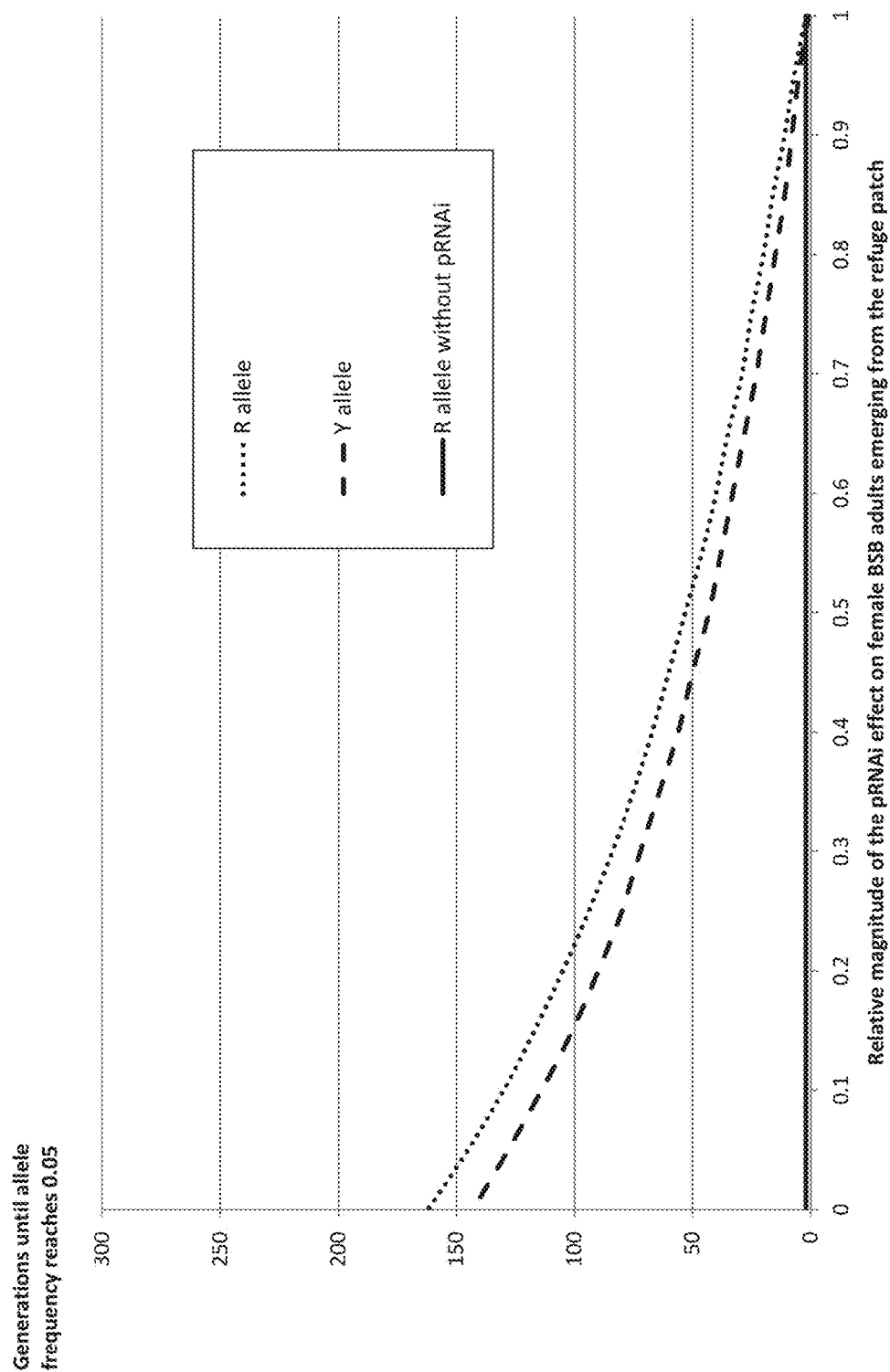


FIG. 8

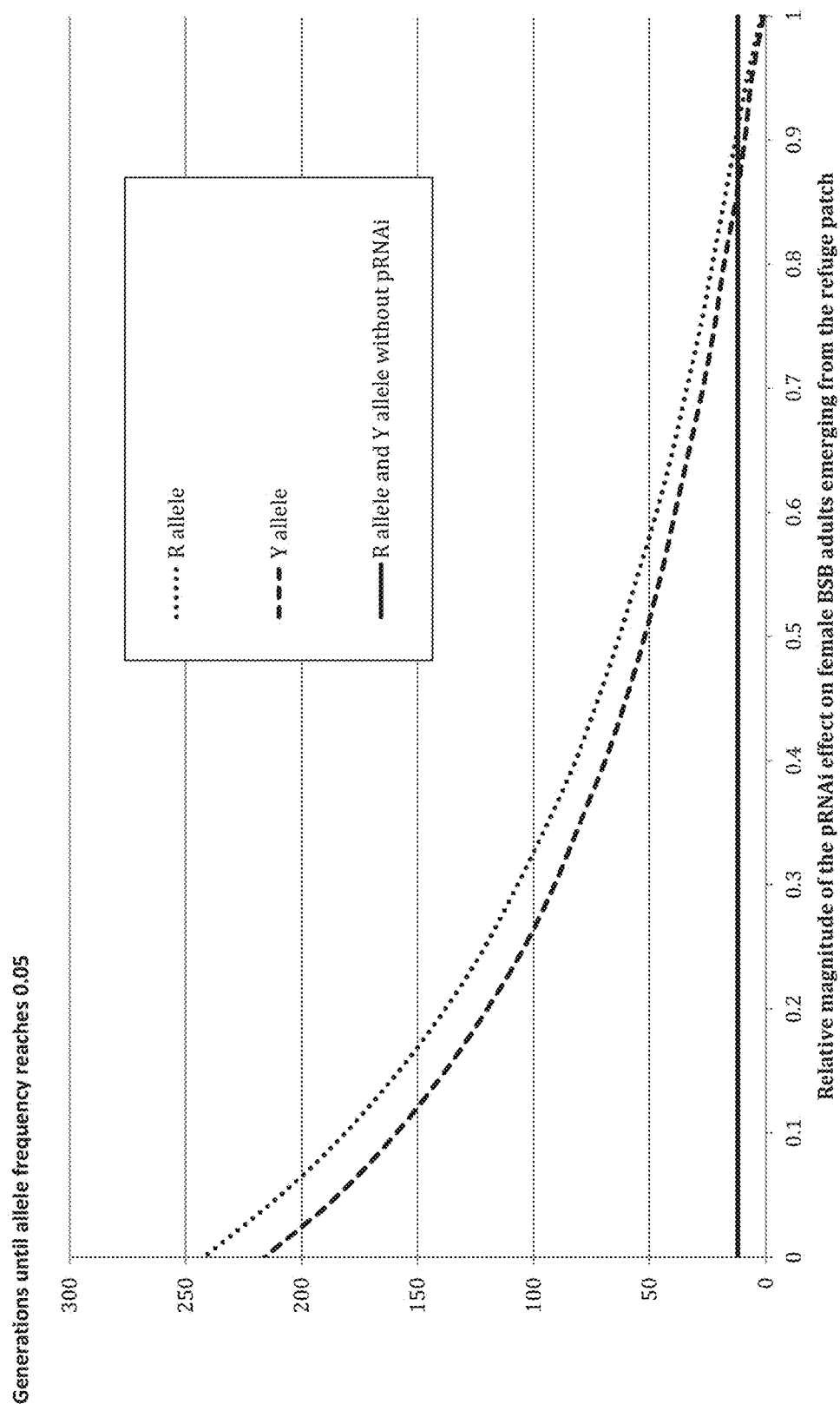


FIG. 9

PARENTAL RNAI SUPPRESSION OF CHROMATIN REMODELING GENES TO CONTROL COLEOPTERAN PESTS

PRIORITY CLAIM

This application claims the benefit of the filing date of U.S. Provisional Patent Application Ser. No. 62/092,747, filed Dec. 16, 2014, the contents of which are incorporated herein in its entirety by this reference.

FIELD OF THE DISCLOSURE

The present invention relates generally to genetic control of plant damage caused by hemipteran pests. In particular embodiments, the present disclosure relates to identification of target coding and non-coding polynucleotides, and the use of recombinant DNA technologies for post-transcriptionally repressing or inhibiting expression of target coding and non-coding polynucleotides in the cells of a hemipteran pest to provide a plant protective effect.

BACKGROUND

Stink bugs and other hemipteran insects (heteroptera) are an important agricultural pest complex. Worldwide, over 50 closely related species of stink bugs are known to cause crop damage. McPherson & McPherson (2000) *Stink bugs of economic importance in America north of Mexico*, CRC Press. Hemipteran insects are present in a large number of important crops including maize, soybean, fruit, vegetables, and cereals.

Stink bugs go through multiple nymph stages before reaching the adult stage. These insects develop from eggs to adults in about 30-40 days. Both nymphs and adults feed on sap from soft tissues into which they also inject digestive enzymes causing extra-oral tissue digestion and necrosis. Digested plant material and nutrients are then ingested. Depletion of water and nutrients from the plant vascular system results in plant tissue damage. Damage to developing grain and seeds is the most significant as yield and germination are significantly reduced. Multiple generations occur in warm climates resulting in significant insect pressure. Current management of stink bugs relies on insecticide treatment on an individual field basis. Therefore, alternative management strategies are urgently needed to minimize ongoing crop losses.

RNA interference (RNAi) is a process utilizing endogenous cellular pathways, whereby an interfering RNA (iRNA) molecule (e.g., a double stranded RNA (dsRNA) molecule) that is specific for all, or any portion of adequate size, of a target gene results in the degradation of the mRNA encoded thereby. In recent years, RNAi has been used to perform gene "knockdown" in a number of species and experimental systems; for example, *Caenorhabditis elegans*, plants, insect embryos, and cells in tissue culture. See, e.g., Fire et al. (1998) Nature 391:806-11; Martinez et al. (2002) Cell 110:563-74; McManus and Sharp (2002) Nature Rev. Genetics 3:737-47.

RNAi accomplishes degradation of mRNA through an endogenous pathway including the DICER protein complex. DICER cleaves long dsRNA molecules into short fragments of approximately 20 nucleotides, termed small interfering RNA (siRNA). The siRNA is unwound into two single-stranded RNAs: the passenger strand and the guide strand. The passenger strand is degraded, and the guide strand is incorporated into the RNA-induced silencing complex

(RISC). Micro inhibitory ribonucleic acids (miRNAs) are structurally very similar molecules that are cleaved from precursor molecules containing a polynucleotide "loop" connecting the hybridized passenger and guide strands, and they may be similarly incorporated into RISC. Post-transcriptional gene silencing occurs when the guide strand binds specifically to a complementary mRNA molecule and induces cleavage by Argonaute, the catalytic component of the RISC complex. This process is known to spread systemically throughout some eukaryotic organisms despite initially limited concentrations of siRNA and/or miRNA such as plants, nematodes, and some insects.

Only transcripts complementary to the siRNA and/or miRNA are cleaved and degraded, and thus the knock-down of mRNA expression is sequence-specific. In plants, several functional groups of DICER genes exist. The gene silencing effect of RNAi persists for days and, under experimental conditions, can lead to a decline in abundance of the targeted transcript of 90% or more, with consequent reduction in levels of the corresponding protein. In insects, there are at least two DICER genes, where DICER1 facilitates miRNA-directed degradation by Argonaute1. Lee et al. (2004) Cell 117 (1):69-81. DICER2 facilitates siRNA-directed degradation by Argonaute2.

The overwhelming majority of sequences complementary to insect DNAs (such as, for example, the 9,000+ sequences identified in U.S. Pat. No. 7,612,194 and U.S. Patent Publication Nos. 2007/0050860, 2010/0192265, and 2011/0154545) do not provide a plant protective effect when used as dsRNA or siRNA. For example, Baum et al. (2007) Nature Biotechnology 25:1322-1326, describe the effects of inhibiting several Western corn rootworm (WCR) gene targets by RNAi. These authors reported that 8 of the 26 target genes they tested were not able to provide experimentally significant coleopteran pest mortality at a very high iRNA (e.g., dsRNA) concentration of more than 520 ng/cm².

The authors of U.S. Pat. No. 7,612,194 and U.S. Patent Publication No. 2007/0050860 made the first report of in planta RNAi in corn plants targeting the western corn rootworm. Baum et al. (2007) Nat. Biotechnol. 25(11):1322-6. These authors describe a high-throughput in vivo dietary RNAi system to screen potential target genes for developing transgenic RNAi maize. Of an initial gene pool of 290 targets, only 14 exhibited larval control potential. One of the most effective double-stranded RNAs (dsRNA) targeted a gene encoding vacuolar ATPase subunit A (V-ATPase), resulting in a rapid suppression of corresponding endogenous mRNA and triggering a specific RNAi response with low concentrations of dsRNA. Thus, these authors documented for the first time the potential for in planta RNAi as a possible pest management tool, while simultaneously demonstrating that effective targets could not be accurately identified a priori, even from a relatively small set of candidate genes.

Another potential application of RNAi for insect control involves parental RNAi (pRNAi). First described in *Caenorhabditis elegans*, pRNAi was identified by injection of dsRNA into the body cavity (or application of dsRNA via ingestion), causing gene inactivity in offspring embryos. Fire et al. (1998), supra; Timmons and Fire (1998) Nature 395(6705):854. A similar process was described in the model coleopteran, *Tribolium castaneum*, whereby female pupae injected with dsRNA corresponding to three unique genes that control segmentation during embryonic development resulted in knock down of zygotic genes in offspring embryos. Bucher et al. (2002) Curr. Biol. 12(3):R85-6. Nearly all of the offspring larvae in this study displayed

gene-specific phenotypes one week after injection. Although injection of dsRNA for functional genomics studies has been successful in a variety of insects, uptake of dsRNA from the gut environment through oral exposure to dsRNA and subsequent down-regulation of essential genes is required in order for RNAi to be effective as a pest management tool. Auer and Frederick (2009) Trends Biotechnol. 27(11):644-51.

Parental RNAi has been used to describe the function of embryonic genes in a number of insect species, including the springtail, *Orchesella cincta* (Konopova and Akam (2014) Evodevo 5(1):2); the brown plant hopper, *Nilaparvata lugens*; the sawfly, *Athalia rosae* (Yoshiyama et al. (2013) J. Insect Physiol. 59(4):400-7); the German cockroach, *Blattella germanica* (Piulachs et al. (2010) Insect Biochem. Mol. Biol. 40:468-75); and the pea aphid, *Acyrtosiphon pisum* (Mao et al. (2013) Arch Insect Biochem Physiol 84(4):209-21). The pRNAi response in all these instances was achieved by injection of dsRNA into the hemocoel of the parental female.

SUMMARY OF THE DISCLOSURE

Disclosed herein are nucleic acid molecules (e.g., target genes, DNAs, dsRNAs, siRNAs, shRNAs, miRNAs, and hpRNAs), and methods of use thereof, for the control of hemipteran pests, including, for example, *Euschistus heros* (Fabr.) (Neotropical Brown Stink Bug, "BSB"); *E. servus* (Say) (Brown Stink Bug); *Nezara viridula* (L.) (Southern Green Stink Bug); *Piezodorus guildinii* (Westwood) (Red-banded Stink Bug); *Halyomorpha halys* (Stål) (Brown Marmorated Stink Bug); *Chinavia hilare* (Say) (Green Stink Bug); *C. marginatum* (Palisot de Beauvois); *Dichelops melacanthus* (Dallas); *D. furcatus* (F.); *Edessa mediatibunda* (F.); *Thyanta perditor* (F.) (Neotropical Red Shouldered Stink Bug); *Horcias nobilellus* (Berg) (Cotton Bug); *Taedia stigmosa* (Berg); *Dysdercus peruvianus* (Guérin-Ménéville); *Neomegalotomus parvus* (Westwood); *Leptoglossus zonatus* (Dallas); *Niesthrea sidae* (F.); *Lygus hesperus* (Knight) (Western Tarnished Plant Bug); and *L. lineolaris* (Palisot de Beauvois). In particular examples, exemplary nucleic acid molecules are disclosed that may be homologous to at least a portion of one or more nucleic acids in a hemipteran pest. In some embodiments, hemipteran pests are controlled by reducing the capacity of an existing generation of the pest to produce a subsequent generation of the pest. In certain examples, delivery of the nucleic acid molecules to hemipteran pests does not result in significant mortality to the pests, but reduces the number of viable progeny produced therefrom.

In these and further examples, the nucleic acid may be a target gene, the product of which may be, for example and without limitation: involved in a metabolic process; involved in a reproductive process; and/or involved in embryonic and/or nymph development. In some examples, post-transcriptional inhibition of the expression of a target gene by a nucleic acid molecule comprising a polynucleotide homologous thereto may result in reduced growth and/or reproduction of the hemipteran pest. In specific examples, a chromatin remodeling gene is selected as a target gene for post-transcriptional silencing. In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_brahma (SEQ ID NO:1 and SEQ ID NO:63). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_mi-2 (SEQ ID NO:8 and

SEQ ID NO:64). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_iswi-1 (SEQ ID NO:10 and SEQ ID NO:65). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_chd1 (SEQ ID NO:14 and SEQ ID NO:67). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_iswi-2 (SEQ ID NO:12 and SEQ ID NO:66). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_ino80 (SEQ ID NO:30). In particular examples, a target gene useful for post-transcriptional inhibition is the novel chromatin remodeling gene referred to herein as BSB_domino (SEQ ID NO:32).

An isolated nucleic acid molecule comprising the polynucleotide of SEQ ID NO:1; the complement of SEQ ID NO:1; SEQ ID NO:8; the complement of SEQ ID NO:8; SEQ ID NO:10; the complement of SEQ ID NO:10; SEQ ID NO:12; the complement of SEQ ID NO:12; SEQ ID NO:14; the complement of SEQ ID NO:14; SEQ ID NO:30; the complement of SEQ ID NO:30; SEQ ID NO:32; the complement of SEQ ID NO:32; SEQ ID NO:63; the complement of SEQ ID NO:63; SEQ ID NO:64; the complement of SEQ ID NO:64; SEQ ID NO:65; the complement of SEQ ID NO:65; SEQ ID NO:66; the complement of SEQ ID NO:66; SEQ ID NO:67; the complement of SEQ ID NO:67; and/or fragments of any of the foregoing (e.g., SEQ ID NO:3, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, and SEQ ID NO:19) is therefore disclosed herein.

Also disclosed are nucleic acid molecules comprising a polynucleotide that encodes a polypeptide that is at least about 85% identical to an amino acid sequence within a target chromatin remodeling gene product (for example, the product of a brahma, mi-2, iswi-1, chd1, iswi-2, ino80, or domino gene). For example, a nucleic acid molecule may comprise a polynucleotide encoding a polypeptide that is at least 85% identical to a polypeptide selected from the group consisting of SEQ ID NO:2 (BSB BRAHMA); an amino acid sequence within a product of BSB brahma; SEQ ID NO:9 (BSB MI-2); an amino acid sequence within a product of BSB mi-2; SEQ ID NO:11 (BSB ISWI-1); an amino acid sequence within a product of BSB iswi-1; SEQ ID NO:15 (BSB CHD1); an amino acid sequence within a product of BSB chd1; SEQ ID NO:13 (BSB ISWI-2); an amino acid sequence within a product of BSB iswi-2; SEQ ID NO:31 (BSB INO80); an amino acid sequence within a product of BSB ino80; SEQ ID NO:33 (BSB DOMINO); and an amino acid sequence within a product of BSB domino. Further disclosed are nucleic acid molecules comprising a polynucleotide that is the reverse complement of a polynucleotide that encodes a polypeptide at least 85% identical to an amino acid sequence within a target chromatin remodeling gene product.

Also disclosed are cDNA polynucleotides that may be used for the production of iRNA (e.g., dsRNA, siRNA, shRNA, miRNA, and hpRNA) molecules that are complementary to all or part of a hemipteran pest target gene, for example, a chromatin remodeling gene. In particular embodiments, dsRNAs, siRNAs, shRNAs, miRNAs, and/or hpRNAs may be produced in vitro or in vivo by a genetically-modified organism, such as a plant or bacterium. In particular examples, cDNA molecules are disclosed that may be used to produce iRNA molecules that are complementary to all or part of mRNA transcribed from BSB_brahma (SEQ ID NO:1 and SEQ ID NO:63), BSB_mi-2

(SEQ ID NO:8 and SEQ ID NO:64), BSB_iswi-1 (SEQ ID NO:10 and SEQ ID NO:65), BSB_chd1 (SEQ ID NO:14 and SEQ ID NO:67), BSB_iswi-2 (SEQ ID NO:12 and SEQ ID NO:66), BSB_ino80 (SEQ ID NO:30), and BSB_domino (SEQ ID NO:32).

Further disclosed are means for inhibiting expression of an essential gene in a hemipteran pest, and means for protecting a plant from a hemipteran pest. A means for inhibiting expression of an essential gene in a hemipteran pest is a single- or double-stranded RNA molecule consisting of a polynucleotide selected from the group consisting of SEQ ID NO:44; SEQ ID NO:49; SEQ ID NO:50; SEQ ID NO:51; SEQ ID NO:52; and the complements thereof. Functional equivalents of means for inhibiting expression of an essential gene in a hemipteran pest include single- or double-stranded RNA molecules that are substantially homologous to all or part of mRNA transcribed from a BSB gene encoding a ATP-dependent remodeling enzyme, such as mRNAs comprising SEQ ID NO:43; SEQ ID NO:45; SEQ ID NO:46; SEQ ID NO:47; SEQ ID NO:48; SEQ ID NO:53; or SEQ ID NO:54. A means for protecting a plant from a hemipteran pest is a DNA molecule comprising a polynucleotide encoding a means for inhibiting expression of an essential gene in a hemipteran pest operably linked to a promoter, wherein the DNA molecule is capable of being integrated into the genome of a soybean plant.

Disclosed are methods for controlling a population of a hemipteran pest, comprising providing to a hemipteran pest an iRNA (e.g., dsRNA, siRNA, shRNA, miRNA, and hpRNA) molecule that functions upon being taken up by the pest to inhibit a biological function within the pest, wherein the iRNA molecule comprises all or part of (e.g., at least 15 contiguous nucleotides of) a polynucleotide selected from the group consisting of: SEQ ID NO:43; the complement of SEQ ID NO:43; SEQ ID NO:44; the complement of SEQ ID NO:44; SEQ ID NO:45; the complement of SEQ ID NO:45; SEQ ID NO:46; the complement of SEQ ID NO:46; SEQ ID NO:47; the complement of SEQ ID NO:47; SEQ ID NO:48; the complement of SEQ ID NO:48; SEQ ID NO:49; the complement of SEQ ID NO:49; SEQ ID NO:50; the complement of SEQ ID NO:50; SEQ ID NO:51; the complement of SEQ ID NO:51; SEQ ID NO:52; the complement of SEQ ID NO:52; SEQ ID NO:53; the complement of SEQ ID NO:53; SEQ ID NO:54; the complement of SEQ ID NO:54; SEQ ID NO:55; the complement of SEQ ID NO:55; SEQ ID NO:56; the complement of SEQ ID NO:56; SEQ ID NO:57; the complement of SEQ ID NO:57; SEQ ID NO:58; the complement of SEQ ID NO:58; SEQ ID NO:59; the complement of SEQ ID NO:59; SEQ ID NO:60; the complement of SEQ ID NO:60; SEQ ID NO:61; the complement of SEQ ID NO:61; SEQ ID NO:62; the complement of SEQ ID NO:62; SEQ ID NO:68; the complement of SEQ ID NO:68; SEQ ID NO:69; the complement of SEQ ID NO:69; SEQ ID NO:70; the complement of SEQ ID NO:70; SEQ ID NO:71; the complement of SEQ ID NO:71; SEQ ID NO:72; the complement of SEQ ID NO:72; a polynucleotide that hybridizes to a coding polynucleotide of a hemipteran organism (e.g., BSB) comprising all or part of any of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; and the complement of a polynucleotide that hybridizes to a coding polynucleotide of a hemipteran organism comprising all or part of any of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67.

Also disclosed herein are methods wherein dsRNAs, siRNAs, shRNAs, miRNAs, and/or hpRNAs may be provided to a hemipteran pest in a diet-based assay, or in genetically-modified plant cells expressing the dsRNAs, siRNAs, shRNAs, miRNAs, and/or hpRNAs. In these and further examples, the dsRNAs, siRNAs, shRNAs, miRNAs, and/or hpRNAs may be ingested by a hemipteran pest. Ingestion of dsRNAs, siRNA, shRNAs, miRNAs, and/or hpRNAs of the invention may then result in RNAi in the pest, which in turn may result in silencing of a gene essential for a metabolic process; a reproductive process; and/or nymph development. Thus, methods are disclosed wherein nucleic acid molecules comprising exemplary polynucleotide(s) useful for parental control of hemipteran pests are provided to a hemipteran pest. In particular examples, the hemipteran pest controlled by use of nucleic acid molecules of the invention may be BSB. In some examples, delivery of the nucleic acid molecules to hemipteran pests does not result in significant mortality to the pests, but reduces the number of viable progeny produced therefrom. In some examples, delivery of the nucleic acid molecules to hemipteran pests results in significant mortality to the pests, and also reduces the number of viable progeny produced therefrom.

The foregoing and other features will become more apparent from the following Detailed Description of several embodiments, which proceeds with reference to the accompanying Figures.

BRIEF DESCRIPTION OF THE FIGURES

FIG. 1A includes a depiction of the strategy used to generate dsRNA from a single transcription template with a single pair of primers, and from two transcription templates (FIG. 1B).

FIG. 2 includes a phylogenetic tree representation of the sequence alignment of ATP-dependent remodelers from *D. v. virgifera* (WCR), *E. heros*, and *Drosophila melanogaster*. For comparison, the tree also contains human BRAHMA, *Saccharomyces cerevisiae* SNF2, and Iswi homologs from the brown marmorated stink bug, *Halyomorpha halys*. The alignment was performed using MUSCLE (100 iterations) in MEGA 6.06. Bootstrap values (MEGA) support the topology of the ATP-dependent remodeler family branches on the maximum likelihood phylogeny tree.

FIGS. 3A-3E includes a representations of the domain architecture of ATP-dependent chromatin remodeling enzymes of *Diabrotica virgifera virgifera* (WCR), *Euschistus heros* (BSB) and *Drosophila melanogaster* (Dme). The graphical representation is of Pfam output, with domains shaded and labeled. The proteins are organized by families and aligned with respect to SNF2 domain. "Squiggly" lines represent truncation/discontinuity for representation purposes.

FIGS. 4A-4C includes data regarding *E. heros* adult female survival, oviposition, and egg hatch rates following dsRNA injections that target chromatin remodeling ATPases. Females were injected with dsRNA at 0 to 2 days post adult molt. FIG. 4A shows the effects on female survival: twenty females were injected with each dsRNA and survival rate was monitored for 23 days. FIG. 4B shows the effects on oviposition: eggs collected from dsRNA-injected females starting at 9 days post-injection. The oviposition rates plotted are per day per female, based on each week of collection. FIG. 4C shows the effects on egg hatching: eggs hatched based on the numbers of eggs laid in

FIG. 4B. Means comparisons were performed with YFP as control using Dunnett's test, † $p < 0.001$, ** $p < 0.05$.

FIG. 5 includes data showing the percent knockdown of chromatin remodeling ATPases in *E. heros* ovaries. Relative expression is represented by $2^{-\Delta\Delta C_t}$. *E. heros* muscle actin transcript was used as a reference gene and ovaries from non-injected females as negative controls. Four sets of ovaries were used in each qRT-PCR experiment. Means comparisons were performed using Student's t-test; † $p < 0.001$.

FIGS. 6A-6B includes data showing the development and hatch rates of eggs oviposited by brahma dsRNA-injected *E. heros* females. Ovipositing females were injected with dsRNA at 14 to 16 days post adult molt. FIG. 6A shows the effects on oviposition: eggs collected from dsRNA-injected females starting at 1 day post-injection. The number of eggs plotted are per day per female, binned into three-day intervals. FIG. 6B shows the effects on egg hatching: eggs hatched based on the numbers in FIG. 6A. Means comparisons were performed with Dunnett's test using non-injected insects as controls, * indicates significance at $p < 0.05$. ** indicates significance at $p < 0.001$.

FIGS. 7A-7H includes data showing the effects on ovaries of *E. heros* females injected with brm or mi-2 dsRNA. FIGS. 7(A-B) show ovaries of non-injected *E. heros* females at zero and four days after adult molt, provided for developmental comparison. FIGS. 7(C-D) show ovaries of females injected with YFP dsRNA, and FIGS. 7(E-F) show brahma dsRNA ovaries at 9 and 14 days post injection. FIG. 7(E) shows lack of ovariole elongation and lack oocyte development, and FIG. 7(F) shows decaying oocytes. FIGS. 7(G-H) show mi-2 dsRNA at 9 and 14 days post injection. FIG. 7(H) shows lack of ovariole elongation, and FIG. 7(G) shows somewhat elongated ovaries with no mature oocytes.

FIG. 8 includes a summary of modeling data showing the relative magnitude of a pRNAi effect on female BSB adults emerging from a "refuge patch" (i.e., that did not express insecticidal iRNAs or recombinant proteins in a transgenic crop). FIG. 8 illustrates the effect on the rate of increase in allele frequencies for resistance to an insecticidal protein (R) and RNAi (Y) when non-refuge plants express the insecticidal protein and parental active iRNA.

FIG. 9 includes a summary of modeling data showing the relative magnitude of a pRNAi effect on female BSB adults emerging from a "refuge patch" (i.e., that did not express insecticidal iRNAs or recombinant proteins in a transgenic crop of plants comprising BSB nymph-active interfering dsRNA in combination with the BSB-active insecticidal protein in the transgenic crop). FIG. 9 illustrates the effect on the rate of increase in allele frequencies for resistance to an insecticidal protein (R) and RNAi (Y) when non-refuge plants express the insecticidal protein and both larval active and parental active iRNA molecules.

SEQUENCE LISTING

The nucleic acid sequences listed in the accompanying sequence listing are shown using standard letter abbreviations for nucleotide bases, as defined in 37 C.F.R. § 1.822. The nucleic acid and amino acid sequences listed define molecules (i.e., polynucleotides and polypeptides, respectively) having the nucleotide and amino acid monomers arranged in the manner described. The nucleic acid and amino acid sequences listed also each define a genus of polynucleotides or polypeptides that comprise the nucleotide and amino acid monomers arranged in the manner described. In view of the redundancy of the genetic code, it

will be understood that a nucleotide sequence including a coding sequence also describes the genus of polynucleotides encoding the same polypeptide as a polynucleotide consisting of the reference sequence. It will further be understood that an amino acid sequence describes the genus of polynucleotide ORFs encoding that polypeptide.

Only one strand of each nucleic acid sequence is shown, but the complementary strand is understood as included by any reference to the displayed strand. As the complement and reverse complement of a primary nucleic acid sequence are necessarily disclosed by the primary sequence, the complementary sequence and reverse complementary sequence of a nucleic acid sequence are included by any reference to the nucleic acid sequence, unless it is explicitly stated to be otherwise (or it is clear to be otherwise from the context in which the sequence appears). Furthermore, as it is understood in the art that the nucleotide sequence of an RNA strand is determined by the sequence of the DNA from which it was transcribed (but for the substitution of uracil (U) nucleobases for thymine (T)), an RNA sequence is included by any reference to the DNA sequence encoding it. In the accompanying sequence listing:

SEQ ID NO:1 shows an exemplary *Euschistus heros* chromatin remodeling gene DNA, referred to herein in some places as brahma.

SEQ ID NO:2 shows the amino acid sequence of a *E. heros* BRAHMA polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:3 shows an exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_brm-1, which is used in some examples for the production of a dsRNA.

SEQ ID NO:4 shows the nucleotide sequence of a T7 phage promoter.

SEQ ID NO:5 shows a segment of an exemplary YFPv2 gene, which is used in some examples for the production of a dsRNA.

SEQ ID NOs:6 and 7 show primers used for PCR amplification of a YFPv2 sequence, used in some examples for dsRNA production.

SEQ ID NO:8 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_mi-2.

SEQ ID NO:9 shows the amino acid sequence of a *E. heros* MI-2 polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:10 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_iswi-1.

SEQ ID NO:11 shows the amino acid sequence of a *E. heros* ISWI-1 polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:12 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_iswi-2.

SEQ ID NO:13 shows the amino acid sequence of a *E. heros* ISWI-2 polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:14 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_chd1.

SEQ ID NO:15 shows the amino acid sequence of a *E. heros* CHD1 polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:16 shows an exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_mi-2-1, which is used in some examples for the production of a dsRNA.

SEQ ID NO:17 shows an exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_iswi-1-1, which is used in some examples for the production of a dsRNA.

SEQ ID NO:18 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_iswi-2-1, which is used in some examples for the production of a dsRNA.

SEQ ID NO:19 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_chd1-1, which is used in some examples for the production of a dsRNA.

SEQ ID NOs:20-29 show primers used to amplify gene regions of chromatin remodeling genes.

SEQ ID NO:30 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_ino80.

SEQ ID NO:31 shows the amino acid sequence of a *E. heros* INO80 polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NO:32 shows a further exemplary *E. heros* chromatin remodeling gene DNA, referred to herein in some places as BSB_domino.

SEQ ID NO:33 shows the amino acid sequence of a *E. heros* DOMINO polypeptide encoded by an exemplary *E. heros* chromatin remodeling gene DNA.

SEQ ID NOs:34-37 show exemplary DNAs encoding dsRNA sequences for targeting SNF2-Helicase regions of insect (e.g., *Euschistus heros*, *Diabrotica*, *Tribolium*, and *Drosophila melanogaster*) chromatin remodeling gene DNA.

SEQ ID NOs:38-41 show exemplary DNAs encoding dsRNA sequences for targeting chromatin remodeling domains (Chromodomain, Bromodomain, or HAND-SLIDE regions) of insect (e.g., *Euschistus heros*, *Diabrotica*, *Tribolium*, and *Drosophila melanogaster*) chromatin remodeling gene DNA.

SEQ ID NO:42 shows an exemplary DNA encoding a YFP v2 hairpin-forming RNA; containing sense polynucleotides, a loop polynucleotide (underlined) including an intron, and antisense polynucleotide (bold font):

ATGTCATCTGGAGCACTTCTCTTTTCATGGGAAGATTCTTACGTTGTGGA
GATGGAAGGGAATGTTGATGGCCACACCTTTAGCATACGTGGGAAAGGCT
ACGGAGATGCCTCAGTGGGAAAGGACTAGTACCGGTTGGGAAAGGTATGT
TTCTGCTTCTACCTTTGATATATATATAATAATTATCACTAATTAGTAGT
AATATAGTATTTCAAGTATTTTTTTCAAAATAAAGAATGTAGTATATAG
CTATTGCTTTTCTGTAGTTTATAAGTGTGTATATTTAATTTATACTTT
TCTAATATATGACCAAAACATGGTGATGTGCGAGTTGATCCGCGGTTACT
TTCCCACTGAGGCATCTCCGTAGCCTTTCCCACTGCTAAAGGTGTGG
CCATCAACATTCCCTTCCATCTCCCAACGTAAGGAATCTTCCCATGAAA
GAGAAGTGCTCCAGATGACAT

SEQ ID NOs:43-62 show exemplary RNAs transcribed from nucleic acids comprising exemplary chromatin remodeling gene polynucleotides and fragments thereof.

SEQ ID NO:63 shows the open reading frame of an exemplary *E. heros* brahma DNA.

SEQ ID NO:64 shows the open reading frame of an exemplary *E. heros* mi-2 DNA.

SEQ ID NO:65 shows the open reading frame of an exemplary *E. heros* iswi-1 DNA.

SEQ ID NO:66 shows the open reading frame of an exemplary *E. heros* iswi-2 DNA.

SEQ ID NO:67 shows the open reading frame of an exemplary *E. heros* chd1 DNA.

SEQ ID NOs:68-72 show further exemplary RNAs transcribed from nucleic acids comprising exemplary chromatin remodeling gene polynucleotides and fragments thereof.

SEQ ID NO:73 shows the open reading frame of an exemplary muscle actin gene.

SEQ ID NOs:74-91 show oligonucleotides and probes used for BSB probe hydrolysis qPCR assay.

DETAILED DESCRIPTION

I. Overview of Several Embodiments

We developed RNA interference (RNAi) as a tool for insect pest management, using a target pest species for transgenic plants that express dsRNA; the Neotropical brown stink bug. Thus far, most genes proposed as targets for RNAi in particular insects do not achieve their purpose, and those useful targets that have been identified involve typically those that cause lethality in the nymph stage. Herein, we describe RNAi-mediated knockdown of chromatin remodeling genes (e.g., brahma, mi-2, chd1, ino80, and domino) in the Neotropical brown stink bug, which is shown to disrupt embryonic development when, for example, iRNA are molecules are delivered via chromatin remodeling gene-targeting dsRNA fed to adult females. There was almost complete absence of hatching in the eggs collected from females exposed to chromatin remodeling gene-targeting dsRNA. In embodiments herein, the ability to deliver chromatin remodeling gene-targeting dsRNA by feeding to adult insects confers a pRNAi effect that is very useful for insect (e.g., hemipteran) pest management. Furthermore, the potential to affect multiple target sequences in both nymph and adult hemipteran pests may increase opportunities to develop sustainable approaches to insect pest management involving RNAi technologies.

Disclosed herein are methods and compositions for genetic control of hemipteran pest infestations. Methods for identifying one or more gene(s) essential to the lifecycle of a hemipteran pest (e.g., gene(s) essential for normal reproductive capacity and/or embryonic and/or nymph development) for use as a target gene for RNAi-mediated control of a hemipteran pest population are also provided. DNA plasmid vectors encoding an RNA molecule may be designed to suppress one or more target gene(s) essential for growth, survival, development, and/or reproduction. In some embodiments, the RNA molecule may be capable of forming dsRNA molecules. In some embodiments, methods are provided for post-transcriptional repression of expression or inhibition of a target gene via nucleic acid molecules that are complementary to a coding or non-coding sequence of the target gene in a hemipteran pest. In these and further embodiments, a hemipteran pest may ingest one or more dsRNA, siRNA, shRNA, miRNA, and/or hpRNA molecules transcribed from all or a portion of a nucleic acid molecule that is complementary to a coding or non-coding sequence of a target gene, thereby providing a plant-protective effect.

Some embodiments involve sequence-specific inhibition of expression of target gene products, using dsRNA, siRNA, shRNA, miRNA and/or hpRNA that is complementary to coding and/or non-coding sequences of the target gene(s) to achieve at least partial control of a hemipteran pest. Disclosed is a set of isolated and purified nucleic acid molecules comprising a polynucleotide, for example, as set forth in SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67; and fragments thereof. In some embodiments, a stabilized dsRNA molecule may be expressed from these polynucleotides, fragments thereof, or a gene comprising one of these polynucleotides, for the post-transcriptional silencing or inhibition of a target gene. In certain embodiments, isolated and purified nucleic acid molecules comprise all or part of any of SEQ ID NO:1; SEQ ID NO:3; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:16; SEQ ID NO:17; SEQ ID NO:18; SEQ ID NO:19; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; and SEQ ID NO:67.

Some embodiments involve a recombinant host cell (e.g., a plant cell) having in its genome at least one recombinant DNA encoding at least one iRNA (e.g., dsRNA) molecule(s). In particular embodiments, the dsRNA molecule(s) may be produced when ingested by a hemipteran pest to post-transcriptionally silence or inhibit the expression of a target gene in the pest or progeny of the pest. The recombinant DNA may comprise, for example, any of SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67; fragments of any of SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67 (e.g., SEQ ID NO:3, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, and SEQ ID NO:19); and a polynucleotide consisting of a partial sequence of a gene comprising one of SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67; fragments of any of SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; SEQ ID NO:67; and/or complements thereof.

Some embodiments involve a recombinant host cell having in its genome a recombinant DNA encoding at least one iRNA (e.g., dsRNA) molecule(s) comprising all or part of SEQ ID NO:43 (e.g., SEQ ID NO:44); all or part of SEQ ID NO:45 (e.g., SEQ ID NO:49); all or part of SEQ ID NO:46 (e.g., SEQ ID NO:50); all or part of SEQ ID NO:47 (e.g., SEQ ID NO:51); all or part of SEQ ID NO:48 (e.g., SEQ ID NO:52); all or part of SEQ ID NO:53; and all or part of SEQ ID NO:54. When ingested by a hemipteran pest, the iRNA molecule(s) may silence or inhibit the expression of a target chromatin remodeling gene (e.g., a DNA comprising all or part of a polynucleotide selected from the group consisting of SEQ ID NO:1; SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:12; SEQ ID NO:14; SEQ ID NO:30; SEQ ID NO:32; SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; and SEQ ID NO:67) in the pest or progeny of the pest, and thereby result in cessation of reproduction in the pest, and/or growth, development, and/or feeding in progeny of the pest.

In some embodiments, a recombinant host cell having in its genome at least one recombinant DNA encoding at least one RNA molecule capable of forming a dsRNA molecule may be a transformed plant cell. Some embodiments involve transgenic plants comprising such a transformed plant cell. In addition to such transgenic plants, progeny plants of any transgenic plant generation, transgenic seeds, and transgenic plant products, are all provided, each of which comprises recombinant DNA(s). In particular embodiments, an RNA molecule capable of forming a dsRNA molecule may be expressed in a transgenic plant cell. Therefore, in these and other embodiments, a dsRNA molecule may be isolated from a transgenic plant cell. In particular embodiments, the transgenic plant is a plant selected from the group comprising corn (*Zea mays*), soybean (*Glycine max*), cotton (*Gossypium* sp.), and plants of the family Poaceae.

Some embodiments involve a method for modulating the expression of a target gene in a hemipteran pest cell. In these and other embodiments, a nucleic acid molecule may be provided, wherein the nucleic acid molecule comprises a polynucleotide encoding an RNA molecule capable of forming a dsRNA molecule. In particular embodiments, a polynucleotide encoding an RNA molecule capable of forming a dsRNA molecule may be operatively linked to a promoter, and may also be operatively linked to a transcription termination sequence. In particular embodiments, a method for modulating the expression of a target gene in a hemipteran pest cell may comprise: (a) transforming a plant cell with a vector comprising a polynucleotide encoding an RNA molecule capable of forming a dsRNA molecule; (b) culturing the transformed plant cell under conditions sufficient to allow for development of a plant cell culture comprising a plurality of transformed plant cells; (c) selecting for a transformed plant cell that has integrated the vector into its genome; and (d) determining that the selected transformed plant cell comprises the RNA molecule capable of forming a dsRNA molecule encoded by the polynucleotide of the vector. A plant may be regenerated from a plant cell that has the vector integrated in its genome and comprises the dsRNA molecule encoded by the polynucleotide of the vector.

Thus, also disclosed is a transgenic plant comprising a vector having a polynucleotide encoding an RNA molecule capable of forming a dsRNA molecule integrated in its genome, wherein the transgenic plant comprises the dsRNA molecule encoded by the polynucleotide of the vector. In particular embodiments, expression of an RNA molecule capable of forming a dsRNA molecule in the plant is sufficient to modulate the expression of a target gene in a cell of a hemipteran pest that contacts the transformed plant or plant cell (for example, by feeding on the transformed plant, a part of the plant (e.g., leaves) or plant cell) or in a cell of a progeny of the hemipteran pest that contacts the transformed plant or plant cell (for example, by parental transmission), such that reproduction of the pest is inhibited. Transgenic plants disclosed herein may display tolerance and/or protection from hemipteran pest infestations. Particular transgenic plants may display protection and/or enhanced protection from one or more pest(s) selected from the group consisting of: *Piezodorus guildinii*; *Halyomorpha halys*; *Nezara viridula*; *Acrosternum hilare*; *Euschistus heros*; *Euschistus servus*; *Chinavia hilare*; *C. marginatum*; *Dichelops melacanthus*; *D. furcatus*; *Edessa mediotabunda*; *Thyanta perditor*; *Horcias nobilellus*; *Taedia stigmosa*; *Dysdercus peruvianus*; *Neomegalotomus parvus*; *Leptoglossus zonatus*; *Niesthrea sidae*; *Lygus hesperus*; and *L. lineolaris*.

Also disclosed herein are methods for delivery of control agents, such as an iRNA molecule, to a hemipteran pest. Such control agents may cause, directly or indirectly, an impairment in the ability of a hemipteran pest population to feed, grow or otherwise cause damage in a host. In some embodiments, a method is provided comprising delivery of a stabilized dsRNA molecule to a hemipteran pest to suppress at least one target gene in the pest or its progeny, thereby causing parental RNAi and reducing or eliminating plant damage in a pest host. In some embodiments, a method of inhibiting expression of a target gene in a hemipteran pest may result in cessation of reproduction in the pest, and/or growth, development, and/or feeding in progeny of the pest. In some embodiments, the method may significantly reduce the size of a subsequent pest generation in an infestation, without directly resulting in mortality in the pest(s) that contact the iRNA molecule. In some embodiments, the method may significantly reduce the size of a subsequent pest generation in an infestation, while also resulting in mortality in the pest(s) that contact the iRNA molecule.

In some embodiments, compositions (e.g., a topical composition) are provided that comprise an iRNA (e.g., dsRNA) molecule for use with plants, animals, and/or the environment of a plant or animal to achieve the elimination or reduction of a hemipteran pest infestation. In particular embodiments, the composition may be a nutritional composition or resource, or food source, to be fed to the hemipteran pest. Some embodiments comprise making the nutritional composition or food source available to the pest. Ingestion of a composition comprising iRNA molecules may result in the uptake of the molecules by one or more cells of the hemipteran pest, which may in turn result in the inhibition of expression of at least one target gene in cell(s) of the pest or its progeny. Ingestion of or damage to a plant or plant cell by a hemipteran pest infestation may be limited or eliminated in or on any host tissue or environment in which the pest is present by providing one or more compositions comprising an iRNA molecule in the host of the pest.

The compositions and methods disclosed herein may be used together in combinations with other methods and compositions for controlling damage by hemipteran pests. For example, an iRNA molecule as described herein for protecting plants from hemipteran pests may be used in a method comprising the additional use of one or more chemical agents effective against a hemipteran pest, biopesticides effective against a hemipteran pest, crop rotation, recombinant genetic techniques that exhibit features different from the features of RNAi-mediated methods and RNAi compositions (e.g., recombinant production of proteins in plants that are harmful to a hemipteran pest (e.g., Bt toxins)), and/or recombinant expression of non-parental iRNA molecules (e.g., lethal iRNA molecules that result in the cessation of growth, development, and/or feeding in the hemipteran pest that contacts the iRNA molecule).

II. Abbreviations

BSB Neotropical brown stink bug (*Euschistus heros*)
dsRNA double-stranded ribonucleic acid
GI growth inhibition
NCBI National Center for Biotechnology Information
gDNA genomic Deoxyribonucleic Acid
iRNA inhibitory ribonucleic acid
ISWI Imitation SWI/imitation switch
ORF open reading frame
RNAi ribonucleic acid interference
miRNA micro ribonucleic acid

siRNA small inhibitory ribonucleic acid
hpRNA hairpin ribonucleic acid
shRNA short hairpin ribonucleic acid
pRNAi parental RNA interference
UTR untranslated region
PCR Polymerase chain reaction
qPCR quantitative polymerase chain reaction
RISC RNA-induced Silencing Complex
RH relative humidity
SEM standard error of the mean
YFP yellow fluorescent protein

III. Terms

In the description and tables which follow, a number of terms are used. In order to provide a clear and consistent understanding of the specification and claims, including the scope to be given such terms, the following definitions are provided:

15 Contact (with an organism): As used herein, the term “contact with” or “uptake by” an organism (e.g., a hemipteran pest), with regard to a nucleic acid molecule, includes internalization of the nucleic acid molecule into the organism, for example and without limitation: ingestion of the molecule by the organism (e.g., by feeding); contacting the organism with a composition comprising the nucleic acid molecule; and soaking of organisms with a solution comprising the nucleic acid molecule.

Contig: As used herein the term “contig” refers to a DNA sequence that is reconstructed from a set of overlapping DNA segments derived from a single genetic source.

Corn plant: As used herein, the term “corn plant” refers to a plant of the species, *Zea mays* (maize). The terms “corn plant” and “maize” are used interchangeably herein.

25 Cotton plant: As used herein, the term “cotton plant” refers to a plant of the species *Gossypium* sp.; for example, *G. hirsutum*.

Expression: As used herein, “expression” of a coding polynucleotide (for example, a gene or a transgene) refers to the process by which the coded information of a nucleic acid transcriptional unit (including, e.g., gDNA or cDNA) is converted into an operational, non-operational, or structural part of a cell, often including the synthesis of a protein. Gene expression can be influenced by external signals; for example, exposure of a cell, tissue, or organism to an agent that increases or decreases gene expression. Expression of a gene can also be regulated anywhere in the pathway from DNA to RNA to protein. Regulation of gene expression occurs, for example, through controls acting on transcription, translation, RNA transport and processing, degradation of intermediary molecules such as mRNA, or through activation, inactivation, compartmentalization, or degradation of specific protein molecules after they have been made, or by combinations thereof. Gene expression can be measured at the RNA level or the protein level by any method known in the art, including, without limitation, northern blot, RT-PCR, western blot, or in vitro, in situ, or in vivo protein activity assay(s).

Genetic material: As used herein, the term “genetic material” includes all genes, and nucleic acid molecules, such as DNA and RNA.

Hemipteran pest: As used herein, the term “hemipteran pest” refers to pest insects of the order Hemiptera, including, for example and without limitation, insects in the families Pentatomidae, Miridae, Pyrrhocoridae, Coreidae, Alydidae, and Rhopalidae, which feed on a wide range of host plants and have piercing and sucking mouth parts. In particular

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examples, a hemipteran pest is selected from the list comprising *Euschistus heros* (Fabr.) (Neotropical Brown Stink Bug), *Nezara viridula* (L.) (Southern Green Stink Bug), *Piezodorus guildinii* (Westwood) (Red-banded Stink Bug), *Halyomorpha halys* (Stål) (Brown Marmorated Stink Bug), *Chinavia hilare* (Say) (Green Stink Bug), *Euschistus servus* (Say) (Brown Stink Bug), *Dichelops melacanthus* (Dallas), *Dichelops furcatus* (F.), *Edessa meditabunda* (F.), *Thyanta perditor* (F.) (Neotropical Red Shouldered Stink Bug), *Chinavia marginatum* (Palisot de Beauvois), *Horcias nobilellus* (Berg) (Cotton Bug), *Taedia stigmosa* (Berg), *Dysdercus peruvianus* (Guérin-Méneville), *Neomegalotomus parvus* (Westwood), *Leptoglossus zonatus* (Dallas), *Niesthrea sidae* (F.), *Lygus hesperus* (Knight) (Western Tarnished Plant Bug), and *Lygus lineolaris* (Palisot de Beauvois).

Inhibition: As used herein, the term “inhibition,” when used to describe an effect on a coding polynucleotide (for example, a gene), refers to a measurable decrease in the cellular level of mRNA transcribed from the coding polynucleotide and/or peptide, polypeptide, or protein product of the coding polynucleotide. In some examples, expression of a coding polynucleotide may be inhibited such that expression is approximately eliminated. “Specific inhibition” refers to the inhibition of a target coding polynucleotide without consequently affecting expression of other coding polynucleotides (e.g., genes) in the cell wherein the specific inhibition is being accomplished.

Isolated: An “isolated” biological component (such as a nucleic acid or protein) has been substantially separated, produced apart from, or purified away from other biological components in the cell of the organism in which the component naturally occurs (i.e., other chromosomal and extra-chromosomal DNA and RNA, and proteins), while effecting a chemical or functional change in the component (e.g., a nucleic acid may be isolated from a chromosome by breaking chemical bonds connecting the nucleic acid to the remaining DNA in the chromosome). Nucleic acid molecules and proteins that have been “isolated” include nucleic acid molecules and proteins purified by standard purification methods. The term also embraces nucleic acids and proteins prepared by recombinant expression in a host cell, as well as chemically-synthesized nucleic acid molecules, proteins, and peptides.

Nucleic acid molecule: As used herein, the term “nucleic acid molecule” may refer to a polymeric form of nucleotides, which may include both sense and anti-sense strands of RNA, cDNA, gDNA, and synthetic forms and mixed polymers of the above. A nucleotide or nucleobase may refer to a ribonucleotide, deoxyribonucleotide, or a modified form of either type of nucleotide. A “nucleic acid molecule” as used herein is synonymous with “nucleic acid” and “polynucleotide.” A nucleic acid molecule is usually at least 10 bases in length, unless otherwise specified. By convention, the nucleotide sequence of a nucleic acid molecule is read from the 5' to the 3' end of the molecule. The “complement” of a nucleic acid molecule refers to a polynucleotide having nucleobases that may form base pairs with the nucleobases of the nucleic acid molecule (i.e., A-T/U, and G-C).

Some embodiments include nucleic acids comprising a template DNA that is transcribed into an RNA molecule that is the complement of an mRNA molecule. In these embodiments, the complement of the nucleic acid transcribed into the mRNA molecule is present in the 5' to 3' orientation, such that RNA polymerase (which transcribes DNA in the 5' to 3' direction) will transcribe a nucleic acid from the complement that can hybridize to the mRNA molecule. Unless explicitly stated otherwise, or it is clear to be

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otherwise from the context, the term “complement” therefore refers to a polynucleotide having nucleobases, from 5' to 3', that may form base pairs with the nucleobases of a reference nucleic acid. Similarly, unless it is explicitly stated to be otherwise (or it is clear to be otherwise from the context), the “reverse complement” of a nucleic acid refers to the complement in reverse orientation. The foregoing is demonstrated in the following illustration:

ATGATGATG polynucleotide

TACTACTAC “complement” of the polynucleotide

CATCATCAT “reverse complement” of the polynucleotide

Some embodiments of the invention may include hairpin RNA-forming RNAi molecules. In these RNAi molecules, both the complement of a nucleic acid to be targeted by RNA interference and the reverse complement may be found in the same molecule, such that the single-stranded RNA molecule may “fold over” and hybridize to itself over region comprising the complementary and reverse complementary polynucleotides.

“Nucleic acid molecules” include all polynucleotides, for example: single- and double-stranded forms of DNA; single-stranded forms of RNA; and double-stranded forms of RNA (dsRNA). The term “nucleotide sequence” or “nucleic acid sequence” refers to both the sense and antisense strands of a nucleic acid as either individual single strands or in the duplex. The term “ribonucleic acid” (RNA) is inclusive of iRNA (inhibitory RNA), dsRNA (double stranded RNA), siRNA (small interfering RNA), shRNA (small hairpin RNA), mRNA (messenger RNA), miRNA (micro-RNA), hpRNA (hairpin RNA), tRNA (transfer RNAs, whether charged or discharged with a corresponding acylated amino acid), and cRNA (complementary RNA). The term “deoxyribonucleic acid” (DNA) is inclusive of cDNA, gDNA, and DNA-RNA hybrids. The terms “polynucleotide” and “nucleic acid,” and “fragments” thereof will be understood by those in the art as a term that includes both gDNAs, ribosomal RNAs, transfer RNAs, messenger RNAs, operons, and smaller engineered polynucleotides that encode or may be adapted to encode, peptides, polypeptides, or proteins.

Oligonucleotide: An oligonucleotide is a short nucleic acid polymer. Oligonucleotides may be formed by cleavage of longer nucleic acid segments, or by polymerizing individual nucleotide precursors. Automated synthesizers allow the synthesis of oligonucleotides up to several hundred bases in length. Because oligonucleotides may bind to a complementary nucleic acid, they may be used as probes for detecting DNA or RNA. Oligonucleotides composed of DNA (oligodeoxyribonucleotides) may be used in PCR, a technique for the amplification of DNAs. In PCR, the oligonucleotide is typically referred to as a “primer,” which allows a DNA polymerase to extend the oligonucleotide and replicate the complementary strand.

A nucleic acid molecule may include either or both naturally occurring and modified nucleotides linked together by naturally occurring and/or non-naturally occurring nucleotide linkages. Nucleic acid molecules may be modified chemically or biochemically, or may contain non-natural or derivatized nucleotide bases, as will be readily appreciated by those of skill in the art. Such modifications include, for example, labels, methylation, substitution of one or more of the naturally occurring nucleotides with an analog, internucleotide modifications (e.g., uncharged linkages: for example, methyl phosphonates, phosphotriesters, phosphoramidates, carbamates, etc.; charged linkages: for example, phosphorothioates, phosphorodithioates, etc.; pen-

dent moieties: for example, peptides; intercalators: for example, acridine, psoralen, etc.; chelators; alkylators; and modified linkages: for example, alpha anomeric nucleic acids, etc.). The term "nucleic acid molecule" also includes any topological conformation, including single-stranded, double-stranded, partially duplexed, triplexed, hairpinned, circular, and padlocked conformations.

As used herein with respect to DNA, the term "coding polynucleotide," "structural polynucleotide," or "structural nucleic acid molecule" refers to a polynucleotide that is ultimately translated into a polypeptide, via transcription and mRNA, when placed under the control of appropriate regulatory elements. With respect to RNA, the term "coding polynucleotide" refers to a polynucleotide that is translated into a peptide, polypeptide, or protein. The boundaries of a coding polynucleotide are determined by a translation start codon at the 5'-terminus and a translation stop codon at the 3'-terminus. Coding polynucleotides include, but are not limited to: gDNA; cDNA; EST; and recombinant polynucleotides.

As used herein, "transcribed non-coding polynucleotide" refers to segments of mRNA molecules such as 5'UTR, 3'UTR and intron segments that are not translated into a peptide, polypeptide, or protein. Further, "transcribed non-coding polynucleotide" refers to a nucleic acid that is transcribed into an RNA that functions in the cell, for example, structural RNAs (e.g., ribosomal RNA (rRNA) as exemplified by 5S rRNA, 5.8S rRNA, 16S rRNA, 18S rRNA, 23S rRNA, and 28S rRNA, and the like); transfer RNA (tRNA); and snRNAs such as U4, U5, U6, and the like. Transcribed non-coding polynucleotides also include, for example and without limitation, small RNAs (sRNA), which term is often used to describe small bacterial non-coding RNAs; small nucleolar RNAs (snoRNA); microRNAs; small interfering RNAs (siRNA); Piwi-interacting RNAs (piRNA); and long non-coding RNAs. Further still, "transcribed non-coding polynucleotide" refers to a polynucleotide that may natively exist as an intragenic "linker" in a nucleic acid and which is transcribed into an RNA molecule.

Lethal RNA interference: As used herein, the term "lethal RNA interference" refers to RNA interference that results in death or a reduction in viability of the subject individual to which, for example, a dsRNA, miRNA, siRNA, shRNA, and/or hpRNA is delivered.

Parental RNA interference: As used herein, the term "parental RNA interference" (pRNAi) refers to a RNA interference phenotype that is observable in progeny of the subject (e.g., a hemipteran pest) to which, for example, a dsRNA, miRNA, siRNA, shRNA, and/or hpRNA is delivered. In some embodiments, pRNAi comprises the delivery of a dsRNA to a hemipteran pest, wherein the pest is thereby rendered less able to produce viable offspring. A nucleic acid that initiates pRNAi may or may not increase the incidence of mortality in a population into which the nucleic acid is delivered. In certain examples, the nucleic acid that initiates pRNAi does not increase the incidence of mortality in the population into which the nucleic acid is delivered. For example, a population of hemipteran pests may be fed one or more nucleic acids that initiate pRNAi, wherein the pests survive and mate but produce eggs that are less able to hatch viable progeny than eggs produced by pests of the same species that are not fed the nucleic acid(s). In one mechanism of pRNAi, parental RNAi delivered to a female is able to knockdown zygotic gene expression in offspring embryos of the female. Bucher et al. (2002) *Curr. Biol.* 12(3):R85-6.

Genome: As used herein, the term "genome" refers to chromosomal DNA found within the nucleus of a cell, and

also refers to organelle DNA found within subcellular components of the cell. In some embodiments of the invention, a DNA molecule may be introduced into a plant cell, such that the DNA molecule is integrated into the genome of the plant cell. In these and further embodiments, the DNA molecule may be either integrated into the nuclear DNA of the plant cell, or integrated into the DNA of the chloroplast or mitochondrion of the plant cell. The term "genome," as it applies to bacteria, refers to both the chromosome and plasmids within the bacterial cell. In some embodiments of the invention, a DNA molecule may be introduced into a bacterium such that the DNA molecule is integrated into the genome of the bacterium. In these and further embodiments, the DNA molecule may be either chromosomally-integrated or located as or in a stable plasmid.

Sequence identity: The term "sequence identity" or "identity," as used herein in the context of two polynucleotides or polypeptides, refers to the residues in the sequences of the two molecules that are the same when aligned for maximum correspondence over a specified comparison window.

As used herein, the term "percentage of sequence identity" may refer to the value determined by comparing two optimally aligned sequences (e.g., nucleic acid sequences or polypeptide sequences) of a molecule over a comparison window, wherein the portion of the sequence in the comparison window may comprise additions or deletions (i.e., gaps) as compared to the reference sequence (which does not comprise additions or deletions) for optimal alignment of the two sequences. The percentage is calculated by determining the number of positions at which the identical nucleotide or amino acid residue occurs in both sequences to yield the number of matched positions, dividing the number of matched positions by the total number of positions in the comparison window, and multiplying the result by 100 to yield the percentage of sequence identity. A sequence that is identical at every position in comparison to a reference sequence is said to be 100% identical to the reference sequence, and vice-versa.

Methods for aligning sequences for comparison are well-known in the art. Various programs and alignment algorithms are described in, for example: Smith and Waterman (1981) *Adv. Appl. Math.* 2:482; Needleman and Wunsch (1970) *J. Mol. Biol.* 48:443; Pearson and Lipman (1988) *Proc. Natl. Acad. Sci. U.S.A.* 85:2444; Higgins and Sharp (1988) *Gene* 73:237-44; Higgins and Sharp (1989) *CABIOS* 5:151-3; Corpet et al. (1988) *Nucleic Acids Res.* 16:10881-90; Huang et al. (1992) *Comp. Appl. Biosci.* 8:155-65; Pearson et al. (1994) *Methods Mol. Biol.* 24:307-31; Tatiana et al. (1999) *FEMS Microbiol. Lett.* 174:247-50. A detailed consideration of sequence alignment methods and homology calculations can be found in, e.g., Altschul et al. (1990) *J. Mol. Biol.* 215:403-10.

The National Center for Biotechnology Information (NCBI) Basic Local Alignment Search Tool (BLAST™; Altschul et al. (1990)) is available from several sources, including the National Center for Biotechnology Information (Bethesda, Md.), and on the internet, for use in connection with several sequence analysis programs. A description of how to determine sequence identity using this program is available on the internet under the "help" section for BLAST™. For comparisons of nucleic acid sequences, the "Blast 2 sequences" function of the BLAST™ (Blastn) program may be employed using the default BLOSUM62 matrix set to default parameters. Nucleic acids with even greater sequence similarity to the sequences of the reference polynucleotides will show increasing percentage identity when assessed by this method.

Specifically hybridizable/Specifically complementary: As used herein, the terms “Specifically hybridizable” and “Specifically complementary” are terms that indicate a sufficient degree of complementarity such that stable and specific binding occurs between the nucleic acid molecule and a target nucleic acid molecule. Hybridization between two nucleic acid molecules involves the formation of an anti-parallel alignment between the nucleobases of the two nucleic acid molecules. The two molecules are then able to form hydrogen bonds with corresponding bases on the opposite strand to form a duplex molecule that, if it is sufficiently stable, is detectable using methods well known in the art. A polynucleotide need not be 100% complementary to its target nucleic acid to be specifically hybridizable. However, the amount of complementarity that must exist for hybridization to be specific is a function of the hybridization conditions used.

Hybridization conditions resulting in particular degrees of stringency will vary depending upon the nature of the hybridization method of choice and the composition and length of the hybridizing nucleic acids. Generally, the temperature of hybridization and the ionic strength (especially the Na⁺ and/or Mg⁺⁺ concentration) of the hybridization buffer will determine the stringency of hybridization, though wash times also influence stringency. Calculations regarding hybridization conditions required for attaining particular degrees of stringency are known to those of ordinary skill in the art, and are discussed, for example, in Sambrook et al. (ed.) *Molecular Cloning: A Laboratory Manual*, 2nd ed., vol. 1-3, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1989, chapters 9 and 11; and Hames and Higgins (eds.) *Nucleic Acid Hybridization*, IRL Press, Oxford, 1985. Further detailed instruction and guidance with regard to the hybridization of nucleic acids may be found, for example, in Tijssen, “Overview of principles of hybridization and the strategy of nucleic acid probe assays,” in *Laboratory Techniques in Biochemistry and Molecular Biology—Hybridization with Nucleic Acid Probes*, Part I, Chapter 2, Elsevier, N.Y., 1993; and Ausubel et al., Eds., *Current Protocols in Molecular Biology*, Chapter 2, Greene Publishing and Wiley-Interscience, NY, 1995.

As used herein, “stringent conditions” encompass conditions under which hybridization will only occur if there is less than 20% mismatch between the sequence of the hybridization molecule and a homologous polynucleotide within the target nucleic acid molecule. “Stringent conditions” include further particular levels of stringency. Thus, as used herein, “moderate stringency” conditions are those under which molecules with more than 20% sequence mismatch will not hybridize; conditions of “high stringency” are those under which sequences with more than 10% mismatch will not hybridize; and conditions of “very high stringency” are those under which sequences with more than 5% mismatch will not hybridize.

The following are representative, non-limiting hybridization conditions.

High Stringency condition (detects polynucleotides that share at least 90% sequence identity): Hybridization in 5×SSC buffer at 65° C. for 16 hours; wash twice in 2×SSC buffer at room temperature for 15 minutes each; and wash twice in 0.5×SSC buffer at 65° C. for 20 minutes each.

Moderate Stringency condition (detects polynucleotides that share at least 80% sequence identity): Hybridization in 5×-6×SSC buffer at 65-70° C. for 16-20 hours; wash twice in 2×SSC buffer at room temperature for 5-20 minutes each; and wash twice in 1×SSC buffer at 55-70° C. for 30 minutes each.

Non-stringent control condition (polynucleotides that share at least 50% sequence identity will hybridize): Hybridization in 6×SSC buffer at room temperature to 55° C. for 16-20 hours; wash at least twice in 2×-3×SSC buffer at room temperature to 55° C. for 20-30 minutes each.

As used herein, the term “substantially homologous” or “substantial homology,” with regard to a nucleic acid, refers to a polynucleotide having contiguous nucleobases that hybridize under stringent conditions to the reference nucleic acid. For example, nucleic acids that are substantially homologous to a reference nucleic acid of any of SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; and SEQ ID NO:67 are those nucleic acids that hybridize under stringent conditions (e.g., the Moderate Stringency conditions set forth, supra) to the reference nucleic acid of SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63; SEQ ID NO:64; SEQ ID NO:65; SEQ ID NO:66; and SEQ ID NO:67. Substantially homologous polynucleotides may have at least 80% sequence identity. For example, substantially homologous polynucleotides may have from about 80% to 100% sequence identity, such as 79%; 80%; about 81%; about 82%; about 83%; about 84%; about 85%; about 86%; about 87%; about 88%; about 89%; about 90%; about 91%; about 92%; about 93%; about 94%; about 95%; about 96%; about 97%; about 98%; about 98.5%; about 99%; about 99.5%; and about 100%. The property of substantial homology is closely related to specific hybridization. For example, a nucleic acid molecule is specifically hybridizable when there is a sufficient degree of complementarity to avoid non-specific binding of the nucleic acid to non-target polynucleotides under conditions where specific binding is desired, for example, under stringent hybridization conditions.

As used herein, the term “ortholog” refers to a gene in two or more species that has evolved from a common ancestral nucleic acid, and may retain the same function in the two or more species.

As used herein, two nucleic acid molecules are said to exhibit “complete complementarity” when every nucleotide of a polynucleotide read in the 5' to 3' direction is complementary to every nucleotide of the other polynucleotide when read in the 3' to 5' direction. A polynucleotide that is complementary to a reference polynucleotide will exhibit a sequence identical to the reverse complement of the reference polynucleotide. These terms and descriptions are well defined in the art and are easily understood by those of ordinary skill in the art.

Operably linked: A first polynucleotide is operably linked with a second polynucleotide when the first polynucleotide is in a functional relationship with the second polynucleotide. When recombinantly produced, operably linked polynucleotides are generally contiguous, and, where necessary to join two protein-coding regions, in the same reading frame (e.g., in a translationally fused ORF). However, nucleic acids need not be contiguous to be operably linked.

The term, “operably linked,” when used in reference to a regulatory genetic element and a coding polynucleotide, means that the regulatory element affects the expression of the linked coding polynucleotide. “Regulatory elements,” or “control elements,” refer to polynucleotides that influence the timing and level/amount of transcription, RNA process-

ing or stability, or translation of the associated coding polynucleotide. Regulatory elements may include promoters; translation leaders; introns; enhancers; stem-loop structures; repressor binding polynucleotides; polynucleotides with a termination sequence; polynucleotides with a polyadenylation recognition sequence; etc. Particular regulatory elements may be located upstream and/or downstream of a coding polynucleotide operably linked thereto. Also, particular regulatory elements operably linked to a coding polynucleotide may be located on the associated complementary strand of a double-stranded nucleic acid molecule.

Promoter: As used herein, the term "promoter" refers to a region of DNA that may be upstream from the start of transcription, and that may be involved in recognition and binding of RNA polymerase and other proteins to initiate transcription. A promoter may be operably linked to a coding polynucleotide for expression in a cell, or a promoter may be operably linked to a polynucleotide encoding a signal peptide which may be operably linked to a coding polynucleotide for expression in a cell. A "plant promoter" may be a promoter capable of initiating transcription in plant cells. Examples of promoters under developmental control include promoters that preferentially initiate transcription in certain tissues, such as leaves, roots, seeds, fibers, xylem vessels, tracheids, or sclerenchyma. Such promoters are referred to as "tissue-preferred". Promoters which initiate transcription only in certain tissues are referred to as "tissue-specific". A "cell type-specific" promoter primarily drives expression in certain cell types in one or more organs, for example, vascular cells in roots or leaves. An "inducible" promoter may be a promoter which may be under environmental control. Examples of environmental conditions that may initiate transcription by inducible promoters include anaerobic conditions and the presence of light. Tissue-specific, tissue-preferred, cell type specific, and inducible promoters constitute the class of "non-constitutive" promoters. A "constitutive" promoter is a promoter which may be active under most environmental conditions or in most tissue or cell types.

Any inducible promoter can be used in some embodiments of the invention. See Ward et al. (1993) Plant Mol. Biol. 22:361-366. With an inducible promoter, the rate of transcription increases in response to an inducing agent. Exemplary inducible promoters include, but are not limited to: Promoters from the ACEI system that respond to copper; In2 gene from maize that responds to benzenesulfonamide herbicide safeners; Tet repressor from Tn10; and the inducible promoter from a steroid hormone gene, the transcriptional activity of which may be induced by a glucocorticosteroid hormone (Schena et al. (1991) Proc. Natl. Acad. Sci. USA 88:0421).

Exemplary constitutive promoters include, but are not limited to: Promoters from plant viruses, such as the 35S promoter from Cauliflower Mosaic Virus (CaMV); promoters from rice actin genes; ubiquitin promoters; pEMU; MAS; maize H3 histone promoter; and the ALS promoter, XbaI/NcoI fragment 5' to the *Brassica napus* ALS3 structural gene (or a polynucleotide similar to said XbaI/NcoI fragment) (International PCT Publication No. WO96/30530).

Additionally, any tissue-specific or tissue-preferred promoter may be utilized in some embodiments of the invention. Plants transformed with a nucleic acid molecule comprising a coding polynucleotide operably linked to a tissue-specific promoter may produce the product of the coding polynucleotide exclusively, or preferentially, in a specific tissue. Exemplary tissue-specific or tissue-preferred promot-

ers include, but are not limited to: A seed-preferred promoter, such as that from the phaseolin gene; a leaf-specific and light-induced promoter such as that from cab or rubisco; an anther-specific promoter such as that from LAT52; a pollen-specific promoter such as that from Zm13; and a microspore-preferred promoter such as that from apg.

Soybean plant: As used herein, the term "soybean plant" refers to a plant of the species *Glycine* sp.; for example, *G. max*.

Transformation: As used herein, the term "transformation" or "transduction" refers to the transfer of one or more nucleic acid molecule(s) into a cell. A cell is "transformed" by a nucleic acid molecule transduced into the cell when the nucleic acid molecule becomes stably replicated by the cell, either by incorporation of the nucleic acid molecule into the cellular genome, or by episomal replication. As used herein, the term "transformation" encompasses all techniques by which a nucleic acid molecule can be introduced into such a cell. Examples include, but are not limited to: transfection with viral vectors; transformation with plasmid vectors; electroporation (Fromm et al. (1986) Nature 319:791-3); lipofection (Felgner et al. (1987) Proc. Natl. Acad. Sci. USA 84:7413-7); microinjection (Mueller et al. (1978) Cell 15:579-85); *Agrobacterium*-mediated transfer (Fraley et al. (1983) Proc. Natl. Acad. Sci. USA 80:4803-7); direct DNA uptake; and microprojectile bombardment (Klein et al. (1987) Nature 327:70).

Transgene: An exogenous nucleic acid. In some examples, a transgene may be a DNA that encodes one or both strand(s) of an RNA capable of forming a dsRNA molecule that comprises a polynucleotide that is complementary to a nucleic acid molecule found in a hemipteran pest. In further examples, a transgene may be an antisense polynucleotide, wherein expression of the antisense polynucleotide inhibits expression of a target nucleic acid, thereby producing a parental RNAi phenotype. In still further examples, a transgene may be a gene (e.g., a herbicide-tolerance gene, a gene encoding an industrially or pharmaceutically useful compound, or a gene encoding a desirable agricultural trait). In these and other examples, a transgene may contain regulatory elements operably linked to a coding polynucleotide of the transgene (e.g., a promoter).

Vector: A nucleic acid molecule as introduced into a cell, for example, to produce a transformed cell. A vector may include genetic elements that permit it to replicate in the host cell, such as an origin of replication. Examples of vectors include, but are not limited to: a plasmid; cosmid; bacteriophage; or virus that carries exogenous DNA into a cell. A vector may also include one or more genes, including ones that produce antisense molecules, and/or selectable marker genes and other genetic elements known in the art. A vector may transduce, transform, or infect a cell, thereby causing the cell to express the nucleic acid molecules and/or proteins encoded by the vector. A vector optionally includes materials to aid in achieving entry of the nucleic acid molecule into the cell (e.g., a liposome, protein coating, etc.).

Yield: A stabilized yield of about 100% or greater relative to the yield of check varieties in the same growing location growing at the same time and under the same conditions. In particular embodiments, "improved yield" or "improving yield" means a cultivar having a stabilized yield of 105% or greater relative to the yield of check varieties in the same growing location containing significant densities of the hemipteran pests that are injurious to that crop growing at the same time and under the same conditions, which are targeted by the compositions and methods herein.

Unless specifically indicated or implied, the terms “a,” “an,” and “the” signify “at least one,” as used herein.

Unless otherwise specifically explained, all technical and scientific terms used herein have the same meaning as commonly understood by those of ordinary skill in the art to which this disclosure belongs. Definitions of common terms in molecular biology can be found in, for example, Lewin's *Genes X*, Jones & Bartlett Publishers, 2009 (ISBN 10 0763766321); Krebs et al. (eds.), *The Encyclopedia of Molecular Biology*, Blackwell Science Ltd., 1994 (ISBN 0-632-02182-9); and Meyers R. A. (ed.), *Molecular Biology and Biotechnology: A Comprehensive Desk Reference*, VCH Publishers, Inc., 1995 (ISBN 1-56081-569-8). All percentages are by weight and all solvent mixture proportions are by volume unless otherwise noted. All temperatures are in degrees Celsius.

IV. Nucleic Acid Molecules Comprising a Hemipteran Pest Polynucleotide

A. Overview

Described herein are nucleic acid molecules useful for the control of hemipteran pests. Described nucleic acid molecules include target polynucleotides (e.g., native genes, and non-coding polynucleotides), dsRNAs, siRNAs, shRNAs, hpRNAs, and miRNAs. For example, dsRNA, siRNA, miRNA, shRNA, and/or hpRNA molecules are described in some embodiments that may be specifically complementary to all or part of one or more nucleic acids in a hemipteran pest. In these and further embodiments, the nucleic acid(s) may be one or more target gene(s), the product of which may be, for example and without limitation: involved in a reproductive process or involved in nymph development. Nucleic acid molecules described herein, when introduced into a cell (e.g., through parental transmission) comprising at least one nucleic acid(s) to which the nucleic acid molecules are specifically complementary, may initiate RNAi in the cell, and consequently reduce or eliminate expression of the native nucleic acid(s). In some examples, reduction or elimination of the expression of a target gene by a nucleic acid molecule specifically complementary thereto may result in reduction or cessation of reproduction in the hemipteran pest, and/or growth, development, and/or feeding in progeny of the pest. These methods may significantly reduce the size of a subsequent pest generation in an infestation, for example, without directly resulting in mortality in the pest(s) that contact the iRNA molecule.

In some embodiments, at least one target gene in a hemipteran pest may be selected, wherein the target gene comprises a chromatin remodeling polynucleotide (e.g., a gene). In particular examples, such a chromatin remodeling gene in a hemipteran pest is selected, wherein the target gene comprises a polynucleotide selected from among BSB_brahma (SEQ ID NO:1 and SEQ ID NO:63); BSB_mi-2 (SEQ ID NO:8 and SEQ ID NO:64); BSB_iswi-1 (SEQ ID NO:10 and SEQ ID NO:65); BSB_chd1 (SEQ ID NO:14 and SEQ ID NO:67); BSB_iswi-2 (SEQ ID NO:12 and SEQ ID NO:66); BSB_ino80 (SEQ ID NO:30); and BSB_domino (SEQ ID NO:32). For example, a target gene in certain embodiments comprises a chromatin remodeling polynucleotide selected from among SEQ ID NO:1, SEQ ID NO: 8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, SEQ ID NO:67; and fragments of any of the foregoing (e.g., SEQ ID NO:3, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, and SEQ ID NO:19).

In some embodiments, a chromatin remodeling polynucleotide encodes a member of the group of “ATP-dependent remodeling enzymes,” a class of ATPases that contain a SNF2 domain (sucrose non-fermenting, originally identified in *Saccharomyces cerevisiae*). ATP-dependent remodeling enzymes include, for example and without limitation, BRAHMA and its orthologs; MI-2 and its orthologs; ISWI, its paralogs, and its orthologs (e.g., ISWI-1 and ISWI-2); CHD1 and its orthologs; INO80 and its orthologs; and DOMINO and its orthologs. Chromatin remodelers (e.g., ATP-dependent remodeling enzymes) exert lasting epigenetic effects by mobilizing nucleosomes and thus changing the access of the transcriptional machinery to DNA.

ATP-dependent remodeling enzymes share the same functional domains and sequence-level conservation. In Pfam (pfam.sanger.ac.uk) searches, ATP-dependent remodeling enzymes can be identified by a combination of SNF2 family N-terminal and Helicase conserved C-terminal (SNF2-Helicase) domains. Thus, RNAi target sites can be designed within the conserved SNF2 family N-terminal and Helicase C-terminal domains (here referred to as SNF2-Helicase) that are common to all chromatin remodelers, as well as chromatin binding or other functional domains that are conserved within each family, which include but are not limited to bromodomain, chromodomain, and HAND-SLIDE domains.

In some embodiments, a target gene may be a nucleic acid molecule comprising a polynucleotide that can be reverse translated in silico to a polypeptide comprising a contiguous amino acid sequence that is at least about 85% identical (e.g., at least 84%, 85%, about 90%, about 95%, about 96%, about 97%, about 98%, about 99%, about 100%, or 100% identical) to the amino acid sequence of a protein product of a chromatin remodeling gene. A target gene may be any nucleic acid in a hemipteran pest, the post-transcriptional inhibition of which has a deleterious effect on the capacity of the pest to produce viable offspring, for example, to provide a protective benefit against the pest to a plant. In particular examples, a target gene is a nucleic acid molecule comprising a polynucleotide that can be reverse translated in silico to a polypeptide comprising a contiguous amino acid sequence that is at least about 85% identical, about 90% identical, about 95% identical, about 96% identical, about 97% identical, about 98% identical, about 99% identical, about 100% identical, or 100% identical to the amino acid sequence that is the in silico translation product of a brahma, mi-2, iswi-1, chd1, iswi-2, ino80, or domino gene. Examples of such translation products include, for example and without limitation: SEQ ID NO:2; SEQ ID NO:9; SEQ ID NO:11; SEQ ID NO:13; SEQ ID NO:15; SEQ ID NO:31; and SEQ ID NO:33.

Provided in some embodiments are DNAs, the expression of which results in an RNA molecule comprising a polynucleotide that is specifically complementary to all or part of a RNA molecule that is encoded by a coding polynucleotide in a hemipteran pest. In some embodiments, after ingestion of the expressed RNA molecule by a hemipteran pest, down-regulation of the coding polynucleotide in cells of the pest, or in cells of progeny of the pest, may be obtained. In particular embodiments, down-regulation of the coding polynucleotide in cells of the hemipteran pest may result in reduction or cessation of reproduction and/or proliferation in the pest, and/or growth, development, and/or feeding in progeny of the pest.

In some embodiments, target polynucleotides include transcribed non-coding RNAs, such as 5'UTRs; 3'UTRs; spliced leaders; introns; outtrons (e.g., 5'UTR RNA subse-

quently modified in trans splicing); donatrons (e.g., non-coding RNA required to provide donor sequences for trans splicing); and other non-coding transcribed RNA of target hemipteran pest genes. Such polynucleotides may be derived from both mono-cistronic and poly-cistronic genes.

Thus, also described herein in connection with some embodiments are iRNA molecules (e.g., dsRNAs, siRNAs, miRNAs, shRNAs, and hpRNAs) that comprise at least one polynucleotide that is specifically complementary to all or part of a target nucleic acid in a hemipteran pest. In some embodiments an iRNA molecule may comprise polynucleotide(s) that are complementary to all or part of a plurality of target nucleic acids; for example, 2, 3, 4, 5, 6, 7, 8, 9, 10, or more target nucleic acids. In particular embodiments, an iRNA molecule may be produced in vitro or in vivo by a genetically-modified organism, such as a plant or bacterium. Also disclosed are cDNAs that may be used for the production of dsRNA molecules, siRNA molecules, miRNA molecules, shRNA molecules, and/or hpRNA molecules that are specifically complementary to all or part of a target nucleic acid in a hemipteran pest. Further described are recombinant DNA constructs for use in achieving stable transformation of particular host targets. Transformed host targets may express effective levels of dsRNA, siRNA, miRNA, shRNA, and/or hpRNA molecules from the recombinant DNA constructs. Therefore, also described is a plant transformation vector comprising at least one polynucleotide operably linked to a heterologous promoter functional in a plant cell, wherein expression of the polynucleotide(s) results in an RNA molecule comprising a string of contiguous nucleobases that are specifically complementary to all or part of a target nucleic acid in a hemipteran pest.

In particular examples, nucleic acid molecules useful for the control of hemipteran pests may include: all or part of a nucleic acid isolated from a hemipteran insect (e.g., BSB) comprising a chromatin remodeling gene polynucleotide (e.g., any of SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67); DNAs that when expressed result in an RNA molecule comprising a polynucleotide that is specifically complementary to all or part of a RNA molecule that is encoded by chromatin remodeling gene; iRNA molecules (e.g., dsRNAs, siRNAs, miRNAs, shRNAs, and hpRNAs) that comprise at least one polynucleotide that is specifically complementary to all or part of an RNA molecule encoded by a chromatin remodeling gene; cDNAs that may be used for the production of dsRNA molecules, siRNA molecules, miRNA molecules, shRNA molecules, and/or hpRNA molecules that are specifically complementary to all or part of an RNA molecule encoded by a chromatin remodeling gene; and recombinant DNA constructs for use in achieving stable transformation of particular host targets, wherein a transformed host target comprises one or more of the foregoing nucleic acid molecules.

B. Nucleic Acid Molecules

The present invention provides, inter alia, iRNA (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) molecules that inhibit target gene expression in a cell, tissue, or organ of a hemipteran pest; and DNA molecules capable of being expressed as an iRNA molecule in a cell or microorganism to inhibit target gene expression in a cell, tissue, or organ of a hemipteran pest.

Some embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two,

three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:1; SEQ ID NO:63; the complement of SEQ ID NO:1; the complement of SEQ ID NO:63; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:1 or SEQ ID NO:63 (e.g., SEQ ID NO:3); the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:1 or SEQ ID NO:63; a coding polynucleotide of a hemipteran insect (e.g., BSB) comprising SEQ ID NO:1 or SEQ ID NO:63; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1 or SEQ ID NO:63; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1 or SEQ ID NO:63; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1 or SEQ ID NO:63. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Alternative embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:8; SEQ ID NO:64; the complement of SEQ ID NO:8; the complement of SEQ ID NO:64; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:8 or SEQ ID NO:64 (e.g., SEQ ID NO:16); the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:8 or SEQ ID NO:64; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8 or SEQ ID NO:64; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8 or SEQ ID NO:64; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8 or SEQ ID NO:64; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8 or SEQ ID NO:64. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Particular embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:10; SEQ ID NO:65; the complement of SEQ ID NO:10; the complement of SEQ ID NO:65; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:10 or SEQ ID NO:65 (e.g., SEQ ID NO:17); the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:10 or SEQ ID NO:65; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10 or SEQ ID NO:65; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10 or SEQ ID NO:65; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10 or SEQ ID NO:65; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10 or SEQ ID NO:65. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Some embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group

consisting of: SEQ ID NO:12; SEQ ID NO:66; the complement of SEQ ID NO:12; the complement of SEQ ID NO:66; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:12 or SEQ ID NO:66 (e.g., SEQ ID NO:18); the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:12 or SEQ ID NO:66; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12 or SEQ ID NO:66; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12 or SEQ ID NO:66; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12 or SEQ ID NO:66; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12 or SEQ ID NO:66. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Other embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:14; SEQ ID NO:67; the complement of SEQ ID NO:14; the complement of SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:14 or SEQ ID NO:67 (e.g., SEQ ID NO:19); the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:14 or SEQ ID NO:67; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14 or SEQ ID NO:67; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14 or SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14 or SEQ ID NO:67; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14 or SEQ ID NO:67. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Some embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:30; the complement of SEQ ID NO:30; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:30; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:30; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

Other embodiments of the invention provide an isolated nucleic acid molecule comprising at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:32; the complement of SEQ ID NO:32; a fragment of at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of SEQ ID NO:32; the complement of a fragment of at least 15 contiguous

nucleotides of SEQ ID NO:32; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development, reproduction and/or feeding of the pest.

In some embodiments, an isolated nucleic acid molecule of the invention may comprise at least one (e.g., one, two, three, or more) polynucleotide(s) selected from the group consisting of: SEQ ID NO:43; the complement of SEQ ID NO:43; SEQ ID NO:44; the complement of SEQ ID NO:44; SEQ ID NO:45; the complement of SEQ ID NO:45; SEQ ID NO:46; the complement of SEQ ID NO:46; SEQ ID NO:47; the complement of SEQ ID NO:47; SEQ ID NO:48; the complement of SEQ ID NO:48; SEQ ID NO:49; the complement of SEQ ID NO:49; SEQ ID NO:50; the complement of SEQ ID NO:50; SEQ ID NO:51; the complement of SEQ ID NO:51; SEQ ID NO:52; the complement of SEQ ID NO:52; SEQ ID NO:53; the complement of SEQ ID NO:53; SEQ ID NO:54; the complement of SEQ ID NO:54; SEQ ID NO:55; the complement of SEQ ID NO:55; SEQ ID NO:56; the complement of SEQ ID NO:56; SEQ ID NO:57; the complement of SEQ ID NO:57; SEQ ID NO:58; the complement of SEQ ID NO:58; SEQ ID NO:59; the complement of SEQ ID NO:59; SEQ ID NO:60; the complement of SEQ ID NO:60; SEQ ID NO:61; the complement of SEQ ID NO:61; SEQ ID NO:62; the complement of SEQ ID NO:62; SEQ ID NO:68; the complement of SEQ ID NO:68; SEQ ID NO:69; the complement of SEQ ID NO:69; SEQ ID NO:70; the complement of SEQ ID NO:70; SEQ ID NO:71; the complement of SEQ ID NO:71; SEQ ID NO:72; the complement of SEQ ID NO:72; a polyribonucleotide transcribed in a hemipteran insect from a gene comprising SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67; the complement of a polyribonucleotide transcribed in a hemipteran insect from a gene comprising SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides of a polyribonucleotide transcribed in a hemipteran insect from a gene comprising SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67; and the complement of a fragment of at least 15 contiguous nucleotides of a polyribonucleotide transcribed in a hemipteran insect from a gene comprising SEQ ID NO:1, SEQ ID NO:3, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67. In particular embodiments, contact with or uptake by a hemipteran pest of the isolated polynucleotide inhibits the growth, development,

reproduction and/or feeding of the pest. In some embodiments, contact with or uptake by the insect occurs via feeding on plant material or bait comprising the iRNA. In some embodiments, contact with or uptake by the insect occurs via spraying of a plant comprising the insect with a composition comprising the iRNA.

In some embodiments, a nucleic acid molecule of the invention may comprise at least one (e.g., one, two, three, or more) DNA(s) capable of being expressed as an iRNA molecule in a cell or microorganism to inhibit target gene expression in a cell, tissue, or organ of a hemipteran pest. Such DNA(s) may be operably linked to a promoter that functions in a cell comprising the DNA molecule to initiate or enhance the transcription of the encoded RNA capable of forming a dsRNA molecule(s). In one embodiment, the at least one (e.g., one, two, three, or more) DNA(s) may be derived from the polynucleotide of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67. Derivatives of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67 includes fragments of these polynucleotides. In some embodiments, such a fragment may comprise, for example, at least about 15 contiguous nucleotides of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, or a complement thereof. Thus, such a fragment may comprise, for example, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200 or more contiguous nucleotides of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, or a complement thereof. In some examples, such a fragment may comprise, for example, at least 19 contiguous nucleotides (e.g., 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, or 30 contiguous nucleotides) of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, or a complement thereof.

Some embodiments comprise introducing partially- or fully-stabilized dsRNA molecules into a hemipteran pest to inhibit expression of a target gene in a cell, tissue, or organ of the hemipteran pest. When expressed as an iRNA molecule (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) and taken up by a hemipteran pest, polynucleotides comprising one or more fragments of any of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; and the complements thereof, may cause one or more of death, developmental arrest, growth inhibition, change in sex ratio, reduction in brood size, cessation of infection, and/or cessation of feeding by a hemipteran pest. In particular examples, polynucleotides comprising one or more fragments (e.g., polynucleotides including about 15 to about 300 nucleotides) of any of S SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; and the complements thereof, cause a reduction in

the capacity of an existing generation of the pest to produce a subsequent generation of the pest.

In certain embodiments, dsRNA molecules provided by the invention comprise polynucleotides complementary to a transcript from a target gene comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67, and/or polynucleotides complementary to a fragment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67, the inhibition of which target gene in a hemipteran pest results in the reduction or removal of a polypeptide or polynucleotide agent that is essential for the pest's or the pest's progeny's growth, development, or other biological function. A selected polynucleotide may exhibit from about 80% to about 100% sequence identity to SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, a contiguous fragment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, or the complement of either of the foregoing. For example, a selected polynucleotide may exhibit 79%; 80%; about 81%; about 82%; about 83%; about 84%; about 85%; about 86%; about 87%; about 88%; about 89%; about 90%; about 91%; about 92%; about 93%; about 94%; about 95%; about 96%; about 97%; about 98%; about 98.5%; about 99%; about 99.5%; or about 100% sequence identity to SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, a contiguous fragment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, or the complement of any of the foregoing.

In some embodiments, a DNA molecule capable of being expressed as an iRNA molecule in a cell or microorganism to inhibit target gene expression may comprise a single polynucleotide that is specifically complementary to all or part of a native polynucleotide found in one or more target hemipteran pest species, or the DNA molecule can be constructed as a chimera from a plurality of such specifically complementary polynucleotides.

In some embodiments, a nucleic acid molecule may comprise a first and a second polynucleotide separated by a "linker." A linker may be a region comprising any sequence of nucleotides that facilitates secondary structure formation between the first and second polynucleotides, where this is desired. In one embodiment, the linker is part of a sense or antisense coding polynucleotide for mRNA. The linker may alternatively comprise any combination of nucleotides or homologues thereof that are capable of being linked covalently to a nucleic acid molecule. In some examples, the linker may comprise an intron (e.g., as ST-LS1 intron).

For example, in some embodiments, the DNA molecule may comprise a polynucleotide coding for one or more different RNA molecules, wherein each of the different RNA molecules comprises a first polynucleotide and a second polynucleotide, wherein the first and second polynucleotides are complementary to each other. The first and second polynucleotides may be connected within an RNA molecule by a linker. The linker may constitute part of the first

polynucleotide or the second polynucleotide. Expression of an RNA molecule comprising the first and second nucleotide polynucleotides may lead to the formation of a dsRNA molecule of the present invention, by specific intramolecular base-pairing of the first and second nucleotide polynucleotides. The first polynucleotide or the second polynucleotide may be substantially identical to a polynucleotide native to a hemipteran pest (e.g., a target gene, or transcribed non-coding polynucleotide), a derivative thereof, or a complementary polynucleotide thereto.

dsRNA nucleic acid molecules comprise double strands of polymerized ribonucleotides, and may include modifications to either the phosphate-sugar backbone or the nucleoside. Modifications in RNA structure may be tailored to allow specific inhibition. In one embodiment, dsRNA molecules may be modified through a ubiquitous enzymatic process so that siRNA molecules may be generated. This enzymatic process may utilize an RNase III enzyme, such as DICER in eukaryotes, either in vitro or in vivo. See Elbashir et al. (2001) *Nature* 411:494-8; and Hamilton and Baulcombe (1999) *Science* 286(5441):950-2. DICER or functionally-equivalent RNase III enzymes cleave larger dsRNA strands and/or hpRNA molecules into smaller oligonucleotides (e.g., siRNAs), each of which is about 19-25 nucleotides in length. The siRNA molecules produced by these enzymes have 2 to 3 nucleotide 3' overhangs, and 5' phosphate and 3' hydroxyl termini. The siRNA molecules generated by RNase III enzymes are unwound and separated into single-stranded RNA in the cell. The siRNA molecules then specifically hybridize with RNAs transcribed from a target gene, and both RNA molecules are subsequently degraded by an inherent cellular RNA-degrading mechanism. This process may result in the effective degradation or removal of the RNA encoded by the target gene in the target organism. The outcome is the post-transcriptional silencing of the targeted gene. In some embodiments, siRNA molecules produced by endogenous RNase III enzymes from heterologous nucleic acid molecules may efficiently mediate the down-regulation of target genes in hemipteran pests.

In some embodiments, a nucleic acid molecule of the invention may include at least one non-naturally occurring polynucleotide that can be transcribed into a single-stranded RNA molecule capable of forming a dsRNA molecule in vivo through intermolecular hybridization. Such dsRNAs typically self-assemble, and can be provided in the nutrition source of a hemipteran pest to achieve the post-transcriptional inhibition of a target gene. In these and further embodiments, a nucleic acid molecule of the invention may comprise two different non-naturally occurring polynucleotides, each of which is specifically complementary to a different target gene in a hemipteran pest. When such a nucleic acid molecule is provided as a dsRNA molecule to a hemipteran pest, the dsRNA molecule inhibits the expression of at least two different target genes in the pest.

C. Obtaining Nucleic Acid Molecules

A variety of polynucleotides in hemipteran pests may be used as targets for the design of nucleic acid molecules of the invention, such as iRNAs and DNA molecules encoding iRNAs. Selection of polynucleotides is not, however, a straight-forward process. Only a small number of polynucleotides in the hemipteran pest will be effective targets. For example, it cannot be predicted with certainty whether a particular polynucleotide can be effectively down-regulated by nucleic acid molecules of the invention, or whether down-regulation of a particular polynucleotide will have a detrimental effect on the growth, viability, proliferation, and/or reproduction of the hemipteran pest. The vast major-

ity of pest polynucleotides, such as ESTs isolated therefrom (e.g., the coleopteran pest polynucleotides listed in U.S. Pat. No. 7,612,194), do not have a detrimental effect on the growth, viability, proliferation, and/or reproduction of the pest. Neither is it predictable which of the polynucleotides that may have a detrimental effect on a hemipteran pest are able to be used in recombinant techniques for expressing nucleic acid molecules complementary to such polynucleotides in a host plant and providing the detrimental effect on the pest upon feeding without causing harm to the host plant.

In some embodiments, nucleic acid molecules of the invention (e.g., dsRNA molecules to be provided in the host plant of a hemipteran pest) are selected to target cDNAs that encode proteins or parts of proteins essential for hemipteran pest reproduction and/or development, such as polypeptides involved in metabolic or catabolic biochemical pathways, cell division, reproduction, energy metabolism, embryonic development, nymph development, transcriptional regulation, and the like. As described herein, contact of compositions by a target organism containing one or more dsRNAs, at least one segment of which is specifically complementary to at least a substantially identical segment of RNA produced in the cells of the target pest organism, can result in failure or reduction of the capacity to mate, lay eggs, or produce viable progeny. A polynucleotide, either DNA or RNA, derived from a hemipteran pest can be used to construct plant cells resistant to infestation by the pests. The host plant of the hemipteran pest (e.g., *Z. mays* or *G. max*), for example, can be transformed to contain one or more of the polynucleotides derived from the hemipteran pest as provided herein. The polynucleotide transformed into the host may encode one or more RNAs that form into a dsRNA structure in the cells or biological fluids within the transformed host, thus making the dsRNA available if/when the pest forms a nutritional relationship with the transgenic host. This may result in the suppression of expression of one or more genes in the cells of the pest, and ultimately inhibition of reproduction and/or development.

Thus, in some embodiments, a gene is targeted that is essentially involved in the growth, development and reproduction of a hemipteran pest. Other target genes for use in the present invention may include, for example, those that play important roles in hemipteran pest viability, movement, migration, growth, development, infectivity, and establishment of feeding sites. A target gene may therefore be a housekeeping gene or a transcription factor. Additionally, a hemipteran pest polynucleotide for use in the present invention may also be derived from a homolog (e.g., an ortholog), of a plant, viral, bacterial or insect gene, the function of which is known to those of skill in the art, and the polynucleotide of which is specifically hybridizable with a target gene in the genome of the target hemipteran pest. Methods of identifying a homolog of a gene with a known nucleotide sequence by hybridization are known to those of skill in the art.

In some embodiments, the invention provides methods for obtaining a nucleic acid molecule comprising a polynucleotide for producing an iRNA (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) molecule. One such embodiment comprises: (a) analyzing one or more target gene(s) for their expression, function, and phenotype upon dsRNA-mediated gene suppression in a hemipteran pest; (b) probing a cDNA or gDNA library with a probe comprising all or a portion of a polynucleotide or a homolog thereof from a targeted hemipteran pest that displays an altered (e.g., reduced) reproduction or development phenotype in a dsRNA-mediated suppression analysis; (c) identifying a DNA clone that

specifically hybridizes with the probe; (d) isolating the DNA clone identified in step (b); (e) sequencing the cDNA or gDNA fragment that comprises the clone isolated in step (d), wherein the sequenced nucleic acid molecule comprises all or a substantial portion of the RNA or a homolog thereof; and (f) chemically synthesizing all or a substantial portion of a gene, or an siRNA, miRNA, hpRNA, mRNA, shRNA, or dsRNA.

In further embodiments, a method for obtaining a nucleic acid fragment comprising a polynucleotide for producing a substantial portion of an iRNA (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) molecule includes: (a) synthesizing first and second oligonucleotide primers specifically complementary to a portion of a polynucleotide from a targeted hemipteran pest; and (b) amplifying a cDNA or gDNA insert present in a cloning vector using the first and second oligonucleotide primers of step (a), wherein the amplified nucleic acid molecule comprises a substantial portion of a siRNA, miRNA, hpRNA, mRNA, shRNA, or dsRNA molecule.

Nucleic acids of the invention can be isolated, amplified, or produced by a number of approaches. For example, an iRNA (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) molecule may be obtained by PCR amplification of a target polynucleotide (e.g., a target gene or a target transcribed non-coding polynucleotide) derived from a gDNA or cDNA library, or portions thereof. DNA or RNA may be extracted from a target organism, and nucleic acid libraries may be prepared therefrom using methods known to those ordinarily skilled in the art. gDNA or cDNA libraries generated from a target organism may be used for PCR amplification and sequencing of target genes. A confirmed PCR product may be used as a template for in vitro transcription to generate sense and antisense RNA with minimal promoters. Alternatively, nucleic acid molecules may be synthesized by any of a number of techniques (See, e.g., Ozaki et al. (1992) *Nucleic Acids Research*, 20: 5205-5214; and Agrawal et al. (1990) *Nucleic Acids Research*, 18: 5419-5423), including use of an automated DNA synthesizer (for example, a P.E. Biosystems, Inc. (Foster City, Calif.) model 392 or 394 DNA/RNA Synthesizer), using standard chemistries, such as phosphoramidite chemistry. See, e.g., Beaucage et al. (1992) *Tetrahedron*, 48: 2223-2311; U.S. Pat. Nos. 4,980,460, 4,725,677, 4,415,732, 4,458,066, and 4,973,679. Alternative chemistries resulting in non-natural backbone groups, such as phosphorothioate, phosphoramidate, and the like, can also be employed.

An RNA, dsRNA, siRNA, miRNA, shRNA, or hpRNA molecule of the present invention may be produced chemically or enzymatically by one skilled in the art through manual or automated reactions, or in vivo in a cell comprising a nucleic acid molecule comprising a polynucleotide encoding the RNA, dsRNA, siRNA, miRNA, shRNA, or hpRNA molecule. RNA may also be produced by partial or total organic synthesis; any modified ribonucleotide can be introduced by in vitro enzymatic or organic synthesis. An RNA molecule may be synthesized by a cellular RNA polymerase or a bacteriophage RNA polymerase (e.g., T3 RNA polymerase, T7 RNA polymerase, and SP6 RNA polymerase). Expression constructs useful for the cloning and expression of polynucleotides are known in the art. See, e.g., International PCT Publication No. WO97/32016; and U.S. Pat. Nos. 5,593,874, 5,698,425, 5,712,135, 5,789,214, and 5,804,693. RNA molecules that are synthesized chemically or by in vitro enzymatic synthesis may be purified prior to introduction into a cell. For example, RNA molecules can be purified from a mixture by extraction with a solvent or

resin, precipitation, electrophoresis, chromatography, or a combination thereof. Alternatively, RNA molecules that are synthesized chemically or by in vitro enzymatic synthesis may be used with no or a minimum of purification, for example, to avoid losses due to sample processing. The RNA molecules may be dried for storage or dissolved in an aqueous solution. The solution may contain buffers or salts to promote annealing, and/or stabilization of dsRNA molecule duplex strands.

In embodiments, a dsRNA molecule may be formed by a single self-complementary RNA strand or from two complementary RNA strands. dsRNA molecules may be synthesized either in vivo or in vitro. An endogenous RNA polymerase of the cell may mediate transcription of the one or two RNA strands in vivo, or cloned RNA polymerase may be used to mediate transcription in vivo or in vitro. Post-transcriptional inhibition of a target gene in a hemipteran pest may be host-targeted by specific transcription in an organ, tissue, or cell type of the host (e.g., by using a tissue-specific promoter); stimulation of an environmental condition in the host (e.g., by using an inducible promoter that is responsive to infection, stress, temperature, and/or chemical inducers); and/or engineering transcription at a developmental stage or age of the host (e.g., by using a developmental stage-specific promoter). RNA strands that form a dsRNA molecule, whether transcribed in vitro or in vivo, may or may not be polyadenylated, and may or may not be capable of being translated into a polypeptide by a cell's translational apparatus.

D. Recombinant Vectors and Host Cell Transformation

In some embodiments, the invention also provides a DNA molecule for introduction into a cell (e.g., a bacterial cell, a yeast cell, or a plant cell), wherein the DNA molecule comprises a polynucleotide that, upon expression to RNA and ingestion by a hemipteran pest, achieves suppression of a target gene in a cell, tissue, or organ of the pest. Thus, some embodiments provide a recombinant nucleic acid molecule comprising a polynucleotide capable of being expressed as an iRNA (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) molecule in a plant cell to inhibit target gene expression in a hemipteran pest. In order to initiate or enhance expression, such recombinant nucleic acid molecules may comprise one or more regulatory elements, which regulatory elements may be operably linked to the polynucleotide capable of being expressed as an iRNA. Methods to express a gene suppression molecule in plants are known, and may be used to express a polynucleotide of the present invention. See, e.g., International PCT Publication No. WO06/073727; and U.S. Patent Publication No. 2006/0200878 A1).

In specific embodiments, a recombinant DNA molecule of the invention may comprise a polynucleotide encoding an RNA that may form a dsRNA molecule. Such recombinant DNA molecules may encode RNAs that may form dsRNA molecules capable of inhibiting the expression of endogenous target gene(s) in a hemipteran pest cell upon ingestion. In many embodiments, a transcribed RNA may form a dsRNA molecule that may be provided in a stabilized form; e.g., as a hairpin and stem and loop structure.

In alternative embodiments, one strand of a dsRNA molecule may be formed by transcription from a polynucleotide which is substantially homologous to the RNA encoded by a polynucleotide selected from the group consisting of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; the complement of

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SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; a coding polynucleotide of a hemipteran insect (e.g., BSB) comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67.

In particular embodiments, a recombinant DNA molecule encoding an RNA that may form a dsRNA molecule may comprise a coding region wherein at least two polynucleotides are arranged such that one polynucleotide is in a sense orientation, and the other polynucleotide is in an antisense orientation, relative to at least one promoter, wherein the sense polynucleotide and the antisense polynucleotide are linked or connected by a linker of, for example, from about five (~5) to about one thousand (~1000) nucleotides. The linker may form a loop between the sense and anti sense polynucleotides. The sense polynucleotide or the antisense polynucleotide may be substantially homologous to an RNA encoded by a target gene (e.g., a chromatin remodeling gene comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67) or fragment thereof. In some embodiments, however, a recombinant DNA molecule may encode an RNA that may form a dsRNA molecule without a linker. In embodiments, a sense coding polynucleotide and an antisense coding polynucleotide may be different lengths.

Polynucleotides identified as having a deleterious effect on hemipteran pests or a plant-protective effect with regard to hemipteran pests may be readily incorporated into expressed dsRNA molecules through the creation of appropriate expression cassettes in a recombinant nucleic acid molecule of the invention. For example, such polynucleotides may be expressed as a hairpin with stem and loop structure by taking a first segment corresponding to an RNA encoded by a target gene polynucleotide (e.g., a chromatin remodeling gene comprising SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64,

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SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67, and fragments thereof); linking this polynucleotide to a second segment linker region that is not homologous or complementary to the first segment; and linking this to a third segment, wherein at least a portion of the third segment is substantially complementary to the first segment. Such a construct forms a stem and loop structure by intramolecular base-pairing of the first segment with the third segment, wherein the loop structure forms comprising the second segment. See, e.g., U.S. Patent Publication Nos. 2002/0048814 and 2003/0018993; and International PCT Publication Nos. WO94/01550 and WO98/05770. A dsRNA molecule may be generated, for example, in the form of a double-stranded structure such as a stem-loop structure (e.g., hairpin), whereby production of siRNA targeted for a native hemipteran pest polynucleotide is enhanced by co-expression of a fragment of the targeted gene, for instance on an additional plant expressible cassette, that leads to enhanced siRNA production, or reduces methylation to prevent transcriptional gene silencing of the dsRNA hairpin promoter.

Embodiments of the invention include introduction of a recombinant nucleic acid molecule of the present invention into a plant (i.e., transformation) to achieve hemipteran pest-protective levels of expression of one or more iRNA molecules. A recombinant DNA molecule may, for example, be a vector, such as a linear or a closed circular plasmid. The vector system may be a single vector or plasmid, or two or more vectors or plasmids that together contain the total DNA to be introduced into the genome of a host. In addition, a vector may be an expression vector. Nucleic acids of the invention can, for example, be suitably inserted into a vector under the control of a suitable promoter that functions in one or more hosts to drive expression of a linked coding polynucleotide or other DNA element. Many vectors are available for this purpose, and selection of the appropriate vector will depend mainly on the size of the nucleic acid to be inserted into the vector and the particular host cell to be transformed with the vector. Each vector contains various components depending on its function (e.g., amplification of DNA or expression of DNA) and the particular host cell with which it is compatible.

To impart protection from a hemipteran pest to a transgenic plant, a recombinant DNA may, for example, be transcribed into an iRNA molecule (e.g., an RNA molecule that forms a dsRNA molecule) within the tissues or fluids of the recombinant plant. An iRNA molecule may comprise a polynucleotide that is substantially homologous and specifically hybridizable to a corresponding transcribed polynucleotide within a hemipteran pest that may cause damage to the host plant species. The hemipteran pest may contact the iRNA molecule that is transcribed in cells of the transgenic host plant, for example, by ingesting cells or fluids of the transgenic host plant that comprise the iRNA molecule. Thus, expression of a target gene is suppressed by the iRNA molecule within hemipteran pests that infest the transgenic host plant. In some embodiments, suppression of expression of the target gene in the target hemipteran pest may result in the plant being tolerant to attack by the pest.

In order to enable delivery of iRNA molecules to a hemipteran pest in a nutritional relationship with a plant cell that has been transformed with a recombinant nucleic acid molecule of the invention, expression (i.e., transcription) of iRNA molecules in the plant cell is required. Thus, a recombinant nucleic acid molecule may comprise a polynucleotide of the invention operably linked to one or more regulatory elements, such as a heterologous promoter element that functions in a host cell, such as a bacterial cell

wherein the nucleic acid molecule is to be amplified, and a plant cell wherein the nucleic acid molecule is to be expressed.

Promoters suitable for use in nucleic acid molecules of the invention include those that are inducible, viral, synthetic, or constitutive, all of which are well known in the art. Non-limiting examples describing such promoters include U.S. Pat. No. 6,437,217 (maize RS81 promoter); U.S. Pat. No. 5,641,876 (rice actin promoter); U.S. Pat. No. 6,426,446 (maize RS324 promoter); U.S. Pat. No. 6,429,362 (maize PR-1 promoter); U.S. Pat. No. 6,232,526 (maize A3 promoter); U.S. Pat. No. 6,177,611 (constitutive maize promoters); U.S. Pat. Nos. 5,322,938, 5,352,605, 5,359,142, and 5,530,196 (CaMV 35S promoter); U.S. Pat. No. 6,433,252 (maize L3 oleosin promoter); U.S. Pat. No. 6,429,357 (rice actin 2 promoter, and rice actin 2 intron); U.S. Pat. No. 6,294,714 (light-inducible promoters); U.S. Pat. No. 6,140,078 (salt-inducible promoters); U.S. Pat. No. 6,252,138 (pathogen-inducible promoters); U.S. Pat. No. 6,175,060 (phosphorous deficiency-inducible promoters); U.S. Pat. No. 6,388,170 (bidirectional promoters); U.S. Pat. No. 6,635,806 (gamma-coixin promoter); and U.S. Patent Publication No. 2009/757,089 (maize chloroplast aldolase promoter). Additional promoters include the nopaline synthase (NOS) promoter (Ebert et al. (1987) Proc. Natl. Acad. Sci. USA 84(16):5745-9) and the octopine synthase (OCS) promoters (which are carried on tumor-inducing plasmids of *Agrobacterium tumefaciens*); the caulimovirus promoters such as the cauliflower mosaic virus (CaMV) 19S promoter (Lawton et al. (1987) Plant Mol. Biol. 9:315-24); the CaMV 35S promoter (Odell et al. (1985) Nature 313:810-2); the figwort mosaic virus 35S-promoter (Walker et al. (1987) Proc. Natl. Acad. Sci. USA 84(19):6624-8); the sucrose synthase promoter (Yang and Russell (1990) Proc. Natl. Acad. Sci. USA 87:4144-8); the R gene complex promoter (Chandler et al. (1989) Plant Cell 1:1175-83); the chlorophyll a/b binding protein gene promoter; CaMV 35S (U.S. Pat. Nos. 5,322,938, 5,352,605, 5,359,142, and 5,530,196); FMV 35S (U.S. Pat. Nos. 6,051,753, and 5,378,619); a PC1SV promoter (U.S. Pat. No. 5,850,019); the SCP1 promoter (U.S. Pat. No. 6,677,503); and AGRtu.nos promoters (GenBank™ Accession No. V00087; Depicker et al. (1982) J. Mol. Appl. Genet. 1:561-73; Bevan et al. (1983) Nature 304:184-7).

In particular embodiments, nucleic acid molecules of the invention comprise a tissue-specific promoter, such as a leaf-specific promoter or pollen-specific promoter. In some embodiments, a polynucleotide or fragment for hemipteran pest control according to the invention may be cloned between two tissue-specific promoters oriented in opposite transcriptional directions relative to the polynucleotide or fragment, and which are operable in a transgenic plant cell and expressed therein to produce RNA molecules in the transgenic plant cell that subsequently may form dsRNA molecules, as described, supra. The iRNA molecules expressed in plant tissues may be ingested by a hemipteran pest so that suppression of target gene expression is achieved.

Additional regulatory elements that may optionally be operably linked to a nucleic acid molecule of interest include 5'UTRs located between a promoter element and a coding polynucleotide that function as a translation leader element. The translation leader element is present in the fully-processed mRNA, and it may affect processing of the primary transcript, and/or RNA stability. Examples of translation leader elements include maize and petunia heat shock protein leaders (U.S. Pat. No. 5,362,865), plant virus coat

protein leaders, plant rubisco leaders, and others. See, e.g., Turner and Foster (1995) Molecular Biotech. 3(3):225-36. Non-limiting examples of 5'UTRs include GmHsp (U.S. Pat. No. 5,659,122); PhDnaK (U.S. Pat. No. 5,362,865); AtAnt1; TEV (Carrington and Freed (1990) J. Virol. 64:1590-7); and AGRtu.nos (GenBank™ Accession No. V00087; and Bevan et al. (1983) Nature 304:184-7).

Additional regulatory elements that may optionally be operably linked to a nucleic acid molecule of interest also include 3' non-translated elements, 3' transcription termination regions, or polyadenylation regions. These are genetic elements located downstream of a polynucleotide, and include polynucleotides that provide polyadenylation signal, and/or other regulatory signals capable of affecting transcription or mRNA processing. The polyadenylation signal functions in plants to cause the addition of polyadenylate nucleotides to the 3' end of the mRNA precursor. The polyadenylation element can be derived from a variety of plant genes, or from T-DNA genes. A non-limiting example of a 3' transcription termination region is the nopaline synthase 3' region (nos 3; Fraley et al. (1983) Proc. Natl. Acad. Sci. USA 80:4803-7). An example of the use of different 3' nontranslated regions is provided in Ingelbrecht et al., (1989) Plant Cell 1:671-80. Non-limiting examples of polyadenylation signals include one from a *Pisum sativum* RbcS2 gene (Ps.RbcS2-E9; Coruzzi et al. (1984) EMBO J. 3:1671-9) and AGRtu.nos (GenBank™ Accession No. E01312).

Some embodiments may include a plant transformation vector that comprises an isolated and purified DNA molecule comprising at least one of the above-described regulatory elements operatively linked to one or more polynucleotides of the present invention. When expressed, the one or more polynucleotides result in one or more RNA molecule(s) comprising a polynucleotide that is specifically complementary to all or part of a RNA molecule in a hemipteran pest. Thus, the polynucleotide(s) may comprise a segment encoding all or part of a polyribonucleotide present within a targeted hemipteran pest RNA transcript, and may comprise inverted repeats of all or a part of a targeted pest transcript. A plant transformation vector may contain polynucleotides specifically complementary to more than one target polynucleotide, thus allowing production of more than one dsRNA for inhibiting expression of two or more genes in cells of one or more populations or species of target hemipteran pests. Segments of polynucleotides specifically complementary to polynucleotides present in different genes can be combined into a single composite nucleic acid molecule for expression in a transgenic plant. Such segments may be contiguous or separated by a linker.

In some embodiments, a plasmid of the present invention already containing at least one polynucleotide(s) of the invention can be modified by the sequential insertion of additional polynucleotide(s) in the same plasmid, wherein the additional polynucleotide(s) are operably linked to the same regulatory elements as the original at least one polynucleotide(s). In some embodiments, a nucleic acid molecule may be designed for the inhibition of multiple target genes. In some embodiments, the multiple genes to be inhibited can be obtained from the same hemipteran pest species, which may enhance the effectiveness of the nucleic acid molecule. In other embodiments, the genes can be derived from a different insect (e.g., hemipteran) pests, which may broaden the range of pests against which the agent(s) is/are effective. When multiple genes are targeted for suppression or a combination of expression and suppression, a polycistronic DNA element can be engineered.

A recombinant nucleic acid molecule or vector of the present invention may comprise a selectable marker that confers a selectable phenotype on a transformed cell, such as a plant cell. Selectable markers may also be used to select for plants or plant cells that comprise a recombinant nucleic acid molecule of the invention. The marker may encode biocide resistance, antibiotic resistance (e.g., kanamycin, Geneticin (G418), bleomycin, hygromycin, etc.), or herbicide tolerance (e.g., glyphosate, etc.). Examples of selectable markers include, but are not limited to: a neo gene which codes for kanamycin resistance and can be selected for using kanamycin, G418, etc.; a bar gene which codes for bialaphos resistance; a mutant EPSP synthase gene which encodes glyphosate tolerance; a nitrilase gene which confers resistance to bromoxynil; a mutant acetolactate synthase (ALS) gene which confers imidazolinone or sulfonylurea resistance; and a methotrexate resistant DHFR gene. Multiple selectable markers are available that confer resistance to ampicillin, bleomycin, chloramphenicol, gentamycin, hygromycin, kanamycin, lincomycin, methotrexate, phosphinothricin, puromycin, spectinomycin, rifampicin, streptomycin and tetracycline, and the like. Examples of such selectable markers are illustrated in, e.g., U.S. Pat. Nos. 5,550,318; 5,633,435; 5,780,708; and 6,118,047.

A recombinant nucleic acid molecule or vector of the present invention may also include a screenable marker. Screenable markers may be used to monitor expression. Exemplary screenable markers include a β -glucuronidase or uidA gene (GUS) which encodes an enzyme for which various chromogenic substrates are known (Jefferson et al. (1987) Plant Mol. Biol. Rep. 5:387-405); an R-locus gene, which encodes a product that regulates the production of anthocyanin pigments (red color) in plant tissues (Dellaporta et al. (1988) "Molecular cloning of the maize R-nj allele by transposon tagging with Ac." In 18th Stadler Genetics Symposium, P. Gustafson and R. Appels, eds. (New York: Plenum), pp. 263-82); a β -lactamase gene (Sutcliffe et al. (1978) Proc. Natl. Acad. Sci. USA 75:3737-41); a gene which encodes an enzyme for which various chromogenic substrates are known (e.g., PADAC, a chromogenic cephalosporin); a luciferase gene (Ow et al. (1986) Science 234:856-9); an xylE gene that encodes a catechol dioxygenase that can convert chromogenic catechols (Zukowski et al. (1983) Gene 46(2-3):247-55); an amylase gene (Ikata et al. (1990) Bio/Technol. 8:241-2); a tyrosinase gene which encodes an enzyme capable of oxidizing tyrosine to DOPA and dopaquinone which in turn condenses to melanin (Katz et al. (1983) J. Gen. Microbiol. 129:2703-14); and an α -galactosidase.

In some embodiments, recombinant nucleic acid molecules, as described, supra, may be used in methods for the creation of transgenic plants and expression of heterologous nucleic acids in plants to prepare transgenic plants that exhibit reduced susceptibility to hemipteran pests. Plant transformation vectors can be prepared, for example, by inserting nucleic acid molecules encoding iRNA molecules into plant transformation vectors and introducing these into plants.

Suitable methods for transformation of host cells include any method by which DNA can be introduced into a cell, such as by transformation of protoplasts (See, e.g., U.S. Pat. No. 5,508,184), by desiccation/inhibition-mediated DNA uptake (See, e.g., Potrykus et al. (1985) Mol. Gen. Genet. 199:183-8), by electroporation (See, e.g., U.S. Pat. No. 5,384,253), by agitation with silicon carbide fibers (See, e.g., U.S. Pat. Nos. 5,302,523 and 5,464,765), by *Agrobacterium*-mediated transformation (See, e.g., U.S. Pat. Nos.

5,563,055; 5,591,616; 5,693,512; 5,824,877; 5,981,840; and 6,384,301) and by acceleration of DNA-coated particles (See, e.g., U.S. Pat. Nos. 5,015,580; 5,550,318; 5,538,880; 6,160,208; 6,399,861; and 6,403,865), etc. Techniques that are particularly useful for transforming corn are described, for example, in U.S. Pat. Nos. 7,060,876 and 5,591,616; and International PCT Publication WO95/06722. Through the application of techniques such as these, the cells of virtually any species may be stably transformed. In some embodiments, transforming DNA is integrated into the genome of the host cell. In the case of multicellular species, transgenic cells may be regenerated into a transgenic organism. Any of these techniques may be used to produce a transgenic plant, for example, comprising one or more nucleic acids encoding one or more iRNA molecules in the genome of the transgenic plant.

The most widely utilized method for introducing an expression vector into plants is based on the natural transformation system of *Agrobacterium*. *A. tumefaciens* and *A. rhizogenes* are plant pathogenic soil bacteria which genetically transform plant cells. The Ti and Ri plasmids of *A. tumefaciens* and *A. rhizogenes*, respectively, carry genes responsible for genetic transformation of the plant. The Ti (tumor-inducing)-plasmids contain a large segment, known as T-DNA, which is transferred to transformed plants. Another segment of the Ti plasmid, the Vir region, is responsible for T-DNA transfer. The T-DNA region is bordered by terminal repeats. In modified binary vectors, the tumor-inducing genes have been deleted, and the functions of the Vir region are utilized to transfer foreign DNA bordered by the T-DNA border elements. The T-region may also contain a selectable marker for efficient recovery of transgenic cells and plants, and a multiple cloning site for inserting polynucleotides for transfer such as a dsRNA encoding nucleic acid.

Thus, in some embodiments, a plant transformation vector is derived from a Ti plasmid of *A. tumefaciens* (See, e.g., U.S. Pat. Nos. 4,536,475, 4,693,977, 4,886,937, and 5,501,967; and European Patent No. EP 0 122 791) or a Ri plasmid of *A. rhizogenes*. Additional plant transformation vectors include, for example and without limitation, those described by Herrera-Estrella et al. (1983) Nature 303:209-13; Bevan et al. (1983) Nature 304:184-7; Klee et al. (1985) Bio/Technol. 3:637-42; and in European Patent No. EP 0 120 516, and those derived from any of the foregoing. Other bacteria such as *Sinorhizobium*, *Rhizobium*, and *Mesorhizobium* that interact with plants naturally can be modified to mediate gene transfer to a number of diverse plants. These plant-associated symbiotic bacteria can be made competent for gene transfer by acquisition of both a disarmed Ti plasmid and a suitable binary vector.

After providing exogenous DNA to recipient cells, transformed cells are generally identified for further culturing and plant regeneration. In order to improve the ability to identify transformed cells, one may desire to employ a selectable or screenable marker gene, as previously set forth, with the transformation vector used to generate the transformant. In the case where a selectable marker is used, transformed cells are identified within the potentially transformed cell population by exposing the cells to a selective agent or agents. In the case where a screenable marker is used, cells may be screened for the desired marker gene trait.

Cells that survive the exposure to the selective agent, or cells that have been scored positive in a screening assay, may be cultured in media that supports regeneration of plants. In some embodiments, any suitable plant tissue culture media (e.g., MS and N6 media) may be modified by

including further substances, such as growth regulators. Tissue may be maintained on a basic medium with growth regulators until sufficient tissue is available to begin plant regeneration efforts, or following repeated rounds of manual selection, until the morphology of the tissue is suitable for regeneration (e.g., at least 2 weeks), then transferred to media conducive to shoot formation. Cultures are transferred periodically until sufficient shoot formation has occurred. Once shoots are formed, they are transferred to media conducive to root formation. Once sufficient roots are formed, plants can be transferred to soil for further growth and maturation.

To confirm the presence of a nucleic acid molecule of interest (for example, a DNA encoding one or more iRNA molecules that inhibit target gene expression in a hemipteran pest) in the regenerating plants, a variety of assays may be performed. Such assays include, for example: molecular biological assays, such as Southern and northern blotting, PCR, and nucleic acid sequencing; biochemical assays, such as detecting the presence of a protein product, e.g., by immunological means (ELISA and/or western blots) or by enzymatic function; plant part assays, such as leaf or root assays; and analysis of the phenotype of the whole regenerated plant.

Integration events may be analyzed, for example, by PCR amplification using, e.g., oligonucleotide primers specific for a nucleic acid molecule of interest. PCR genotyping is understood to include, but not be limited to, polymerase-chain reaction (PCR) amplification of gDNA derived from isolated host plant callus tissue predicted to contain a nucleic acid molecule of interest integrated into the genome, followed by standard cloning and sequence analysis of PCR amplification products. Methods of PCR genotyping have been well described (for example, Rios, G. et al. (2002) *Plant J.* 32:243-53) and may be applied to gDNA derived from any plant species (e.g., *Z. mays* or *G. max*) or tissue type, including cell cultures.

A transgenic plant formed using *Agrobacterium*-dependent transformation methods typically contains a single recombinant DNA inserted into one chromosome. The polynucleotide of the single recombinant DNA is referred to as a "transgenic event" or "integration event". Such transgenic plants are heterozygous for the inserted exogenous polynucleotide. In some embodiments, a transgenic plant homozygous with respect to a transgene may be obtained by sexually mating (selfing) an independent segregant transgenic plant that contains a single exogenous gene to itself, for example a T₀ plant, to produce T₁ seed. One fourth of the T₁ seed produced will be homozygous with respect to the transgene. Germinating T₁ seed results in plants that can be tested for heterozygosity, typically using an SNP assay or a thermal amplification assay that allows for the distinction between heterozygotes and homozygotes (i.e., a zygosity assay).

In particular embodiments, at least 2, 3, 4, 5, 6, 7, 8, 9 or 10 or more different iRNA molecules are produced in a plant cell that have a hemipteran pest-protective effect. The iRNA molecules (e.g., dsRNA molecules) may be expressed from multiple nucleic acids introduced in different transformation events, or from a single nucleic acid introduced in a single transformation event. In some embodiments, a plurality of iRNA molecules are expressed under the control of a single promoter. In other embodiments, a plurality of iRNA molecules are expressed under the control of multiple promoters. Single iRNA molecules may be expressed that comprise multiple polynucleotides that are each homologous to different loci within one or more hemipteran pests (for

example, the loci defined by SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67), both in different populations of the same species of hemipteran pest, or in different species of hemipteran pests.

In addition to direct transformation of a plant with a recombinant nucleic acid molecule, transgenic plants can be prepared by crossing a first plant having at least one transgenic event with a second plant lacking such an event. For example, a recombinant nucleic acid molecule comprising a polynucleotide that encodes an iRNA molecule may be introduced into a first plant line that is amenable to transformation to produce a transgenic plant, which transgenic plant may be crossed with a second plant line to introgress the polynucleotide that encodes the iRNA molecule into the second plant line.

The invention also includes commodity products containing one or more of the polynucleotides of the present invention. Particular embodiments include commodity products produced from a recombinant plant or seed containing one or more of the polynucleotides of the present invention. A commodity product containing one or more of the polynucleotides of the present invention is intended to include, but not be limited to, meals, oils, crushed or whole grains or seeds of a plant, or any food product comprising any meal, oil, or crushed or whole grain of a recombinant plant or seed containing one or more of the polynucleotides of the present invention. The detection of one or more of the polynucleotides of the present invention in one or more commodity or commodity products contemplated herein is de facto evidence that the commodity or commodity product is produced from a transgenic plant designed to express one or more of the polynucleotides of the present invention for the purpose of controlling plant pests using dsRNA-mediated gene suppression methods.

In some aspects, seeds and commodity products produced by transgenic plants derived from transformed plant cells are included, wherein the seeds or commodity products comprise a detectable amount of a nucleic acid of the invention. In some embodiments, such commodity products may be produced, for example, by obtaining transgenic plants and preparing food or feed from them. Commodity products comprising one or more of the polynucleotides of the invention includes, for example and without limitation: meals, oils, crushed or whole grains or seeds of a plant, and any food product comprising any meal, oil, or crushed or whole grain of a recombinant plant or seed comprising one or more of the nucleic acids of the invention. The detection of one or more of the polynucleotides of the invention in one or more commodity or commodity products is de facto evidence that the commodity or commodity product is produced from a transgenic plant designed to express one or more of the iRNA molecules of the invention for the purpose of controlling hemipteran pests.

In some embodiments, a transgenic plant or seed comprising a nucleic acid molecule of the invention also may comprise at least one other transgenic event in its genome, including without limitation: a transgenic event from which is transcribed an iRNA molecule targeting a locus in a hemipteran pest other than the ones defined by SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67; a transgenic event from which is transcribed an iRNA molecule targeting a gene in an organism other than a hemipteran pest (e.g., a plant-parasitic nematode); a gene

encoding an insecticidal protein (e.g., a *Bacillus thuringiensis* insecticidal protein); an herbicide tolerance gene (e.g., a gene providing tolerance to glyphosate); and a gene contributing to a desirable phenotype in the transgenic plant, such as increased yield, altered fatty acid metabolism, or restoration of cytoplasmic male sterility. In particular embodiments, polynucleotides encoding iRNA molecules of the invention may be combined with other insect control and disease traits in a plant to achieve desired traits for enhanced control of plant disease and insect damage. Combining insect control traits that employ distinct modes-of-action may provide protected transgenic plants with superior durability over plants harboring a single control trait, for example, because of the reduced probability that resistance to the trait(s) will develop in the field.

V. Target Gene Suppression in a Hemipteran Pest

A. Overview

In some embodiments of the invention, at least one nucleic acid molecule useful for the control of hemipteran pests may be provided to a hemipteran pest, wherein the nucleic acid molecule leads to RNAi-mediated gene silencing in the pest(s). In particular embodiments, an iRNA molecule (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) may be provided to the hemipteran host. In some embodiments, a nucleic acid molecule useful for the control of hemipteran pests may be provided to a pest by contacting the nucleic acid molecule with the pest. In these and further embodiments, a nucleic acid molecule useful for the control of hemipteran pests may be provided in a feeding substrate of the pest, for example, a nutritional composition. In these and further embodiments, a nucleic acid molecule useful for the control of a hemipteran pest may be provided through ingestion of plant material comprising the nucleic acid molecule that is ingested by the pest(s). In certain embodiments, the nucleic acid molecule is present in plant material through expression of a recombinant nucleic acid introduced into the plant material, for example, by transformation of a plant cell with a vector comprising the recombinant nucleic acid and regeneration of a plant material or whole plant from the transformed plant cell.

B. RNAi-mediated Target Gene Suppression

In embodiments, the invention provides iRNA molecules (e.g., dsRNA, siRNA, miRNA, shRNA, and hpRNA) that may be designed to target essential polynucleotides (e.g., essential genes) in the transcriptome of a hemipteran (e.g., BSB) pest, for example by designing an iRNA molecule that comprises at least one strand comprising a polynucleotide that is specifically complementary to the target polynucleotide. The sequence of an iRNA molecule so designed may be identical to that of the target polynucleotide, or may incorporate mismatches that do not prevent specific hybridization between the iRNA molecule and its target polynucleotide.

iRNA molecules of the invention may be used in methods for gene suppression in a hemipteran pest, thereby reducing the level or incidence of damage caused by the pest on a plant (for example, a protected transformed plant comprising an iRNA molecule). As used herein the term “gene suppression” refers to any of the well-known methods for reducing the levels of protein produced as a result of gene transcription to mRNA and subsequent translation of the mRNA, including the reduction of protein expression from a gene or a coding polynucleotide including post-transcriptional inhibition of expression and transcriptional suppression. Post-transcriptional inhibition is mediated by specific

homology between all or a part of an mRNA transcribed from a gene targeted for suppression and the corresponding iRNA molecule used for suppression. Additionally, post-transcriptional inhibition refers to the substantial and measurable reduction of the amount of mRNA available in the cell for binding by ribosomes.

In embodiments wherein an iRNA molecule is a dsRNA molecule, the dsRNA molecule may be cleaved by the enzyme, DICER, into short siRNA molecules (approximately 20 nucleotides in length). The double-stranded siRNA molecule generated by DICER activity upon the dsRNA molecule may be separated into two single-stranded siRNAs; the “passenger strand” and the “guide strand”. The passenger strand may be degraded, and the guide strand may be incorporated into RISC. Post-transcriptional inhibition occurs by specific hybridization of the guide strand with a specifically complementary polynucleotide of an mRNA molecule, and subsequent cleavage by the enzyme, Argonaute (catalytic component of the RISC complex).

In embodiments of the invention, any form of iRNA molecule may be used. Those of skill in the art will understand that dsRNA molecules typically are more stable during preparation and during the step of providing the iRNA molecule to a cell than are single-stranded RNA molecules, and are typically also more stable in a cell. Thus, while siRNA and miRNA molecules, for example, may be equally effective in some embodiments, a dsRNA molecule may be chosen due to its stability.

In particular embodiments, a nucleic acid molecule is provided that comprises a polynucleotide, which polynucleotide may be expressed in vitro to produce an iRNA molecule that is substantially homologous to a nucleic acid molecule encoded by a polynucleotide within the genome of a hemipteran pest. In certain embodiments, the in vitro transcribed iRNA molecule may be a stabilized dsRNA molecule that comprises a stem-loop structure. After a hemipteran pest contacts the in vitro transcribed iRNA molecule, post-transcriptional inhibition of a target gene in the pest (for example, an essential gene) may occur.

In some embodiments of the invention, expression of an iRNA from a nucleic acid molecule comprising at least 15 contiguous nucleotides (e.g., at least 19 contiguous nucleotides) of a polynucleotide are used in a method for post-transcriptional inhibition of a target gene in a hemipteran pest, wherein the polynucleotide is selected from the group consisting of: SEQ ID NO:1; the complement of SEQ ID NO:1; SEQ ID NO:8; the complement of SEQ ID NO:8; SEQ ID NO:10; the complement of SEQ ID NO:10; SEQ ID NO:12; the complement of SEQ ID NO:12; SEQ ID NO:14; the complement of SEQ ID NO:14; SEQ ID NO:30; the complement of SEQ ID NO:30; SEQ ID NO:32; the complement of SEQ ID NO:32; SEQ ID NO:63; the complement of SEQ ID NO:63; SEQ ID NO:64; the complement of SEQ ID NO:64; SEQ ID NO:65; the complement of SEQ ID NO:65; SEQ ID NO:66; the complement of SEQ ID NO:66; SEQ ID NO:67; the complement of SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:1; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:1; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:8; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:8; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:10; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:10; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:12; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:12; a fragment of at least 15 contiguous

nucleotides of SEQ ID NO:14; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:14; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:30; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:30; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:32; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:32; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:63; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:63; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:64; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:64; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:65; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:65; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:66; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:66; a fragment of at least 15 contiguous nucleotides of SEQ ID NO:67; the complement of a fragment of at least 15 contiguous nucleotides of SEQ ID NO:67; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:63; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:63; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:64; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:64; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:65; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:65; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:66; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:66; a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:67; the complement of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:67; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:1; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:8; a fragment of at least 15 contiguous nucleotides of a coding

polynucleotide of a hemipteran insect comprising SEQ ID NO:10; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:10; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:12; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:14; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:30; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:32; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:63; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:63; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:64; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:64; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:65; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:65; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:66; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:66; a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:67; and the complement of a fragment of at least 15 contiguous nucleotides of a coding polynucleotide of a hemipteran insect comprising SEQ ID NO:67. In certain embodiments, expression of a nucleic acid molecule that is at least about 80% identical (e.g., 79%, about 80%, about 81%, about 82%, about 83%, about 84%, about 85%, about 86%, about 87%, about 88%, about 89%, about 90%, about 91%, about 92%, about 93%, about 94%, about 95%, about 96%, about 97%, about 98%, about 99%, about 100%, and 100%) with any of the foregoing may be used. In these and further embodiments, a nucleic acid molecule may be expressed that specifically hybridizes to an RNA molecule present in at least one cell of a hemipteran pest.

It is an important feature of some embodiments herein that the RNAi post-transcriptional inhibition system is able to tolerate sequence variations among target genes that might be expected due to genetic mutation, strain polymorphism, or evolutionary divergence. The introduced nucleic acid molecule may not need to be absolutely homologous to either a primary transcription product or a fully-processed mRNA of a target gene, so long as the introduced nucleic acid molecule is specifically hybridizable to either a primary

transcription product or a fully-processed mRNA of the target gene. Moreover, the introduced nucleic acid molecule may not need to be full-length, relative to either a primary transcription product or a fully processed mRNA of the target gene.

Inhibition of a target gene using the iRNA technology of the present invention is sequence-specific; i.e., polynucleotides substantially homologous to the iRNA molecule(s) are targeted for genetic inhibition. In some embodiments, an RNA molecule comprising a polynucleotide with a nucleotide sequence that is identical to that of a portion of a target gene may be used for inhibition. In these and further embodiments, an RNA molecule comprising a polynucleotide with one or more insertion, deletion, and/or point mutations relative to a target polynucleotide may be used. In particular embodiments, an iRNA molecule and a portion of a target gene may share, for example, at least from about 80%, at least from about 81%, at least from about 82%, at least from about 83%, at least from about 84%, at least from about 85%, at least from about 86%, at least from about 87%, at least from about 88%, at least from about 89%, at least from about 90%, at least from about 91%, at least from about 92%, at least from about 93%, at least from about 94%, at least from about 95%, at least from about 96%, at least from about 97%, at least from about 98%, at least from about 99%, at least from about 100%, and 100% sequence identity. Alternatively, the duplex region of a dsRNA molecule may be specifically hybridizable with a portion of a target gene transcript. In specifically hybridizable molecules, a less than full length polynucleotide exhibiting a greater homology compensates for a longer, less homologous polynucleotide. The length of the polynucleotide of a duplex region of a dsRNA molecule that is identical to a portion of a target gene transcript may be at least about 25, 50, 100, 200, 300, 400, 500, or at least about 1000 bases. In some embodiments, a polynucleotide of greater than 20-100 nucleotides may be used; for example, a polynucleotide of 100-200 or 300-500 nucleotides may be used. In particular embodiments, a polynucleotide of greater than about 200-300 nucleotides may be used. In particular embodiments, a polynucleotide of greater than about 500-1000 nucleotides may be used, depending on the size of the target gene.

In certain embodiments, expression of a target gene in a hemipteran pest may be inhibited by at least 10%; at least 33%; at least 50%; or at least 80% within a cell of the pest, such that a significant inhibition takes place. Significant inhibition refers to inhibition over a threshold that results in a detectable phenotype (e.g., cessation of reproduction, feeding, development, etc.), or a detectable decrease in RNA and/or gene product corresponding to the target gene being inhibited. Although in certain embodiments of the invention inhibition occurs in substantially all cells of the pest, in other embodiments inhibition occurs only in a subset of cells expressing the target gene.

In some embodiments, transcriptional suppression is mediated by the presence in a cell of a dsRNA molecule exhibiting substantial sequence identity to a promoter DNA or the complement thereof to effect what is referred to as "promoter trans suppression." Gene suppression may be effective against target genes in a hemipteran pest that may ingest or contact such dsRNA molecules, for example, by ingesting or contacting plant material containing the dsRNA molecules. dsRNA molecules for use in promoter trans suppression may be specifically designed to inhibit or suppress the expression of one or more homologous or complementary polynucleotides in the cells of the hemipteran pest. Post-transcriptional gene suppression by antisense or sense

oriented RNA to regulate gene expression in plant cells is disclosed in U.S. Pat. Nos. 5,107,065; 5,759,829; 5,283,184; and 5,231,020.

C. Expression of iRNA Molecules Provided to a Hemipteran Pest

Expression of iRNA molecules for RNAi-mediated gene inhibition in a hemipteran pest may be carried out in any one of many in vitro or in vivo formats. The iRNA molecules may then be provided to a hemipteran pest, for example, by contacting the iRNA molecules with the pest, or by causing the pest to ingest or otherwise internalize the iRNA molecules. Some embodiments of the invention include transformed host plants of a hemipteran pest, transformed plant cells, and progeny of transformed plants. The transformed plant cells and transformed plants may be engineered to express one or more of the iRNA molecules, for example, under the control of a heterologous promoter, to provide a pest-protective effect. Thus, when a transgenic plant or plant cell is consumed by a hemipteran pest during feeding, the pest may ingest iRNA molecules expressed in the transgenic plants or cells. The polynucleotides of the present invention may also be introduced into a wide variety of prokaryotic and eukaryotic microorganism hosts to produce iRNA molecules. The term "microorganism" includes prokaryotic and eukaryotic species, such as bacteria and fungi.

Modulation of gene expression may include partial or complete suppression of such expression. In another embodiment, a method for suppression of gene expression in a hemipteran pest comprises providing in the tissue of the host of the pest a gene-suppressive amount of at least one dsRNA molecule formed following transcription of a polynucleotide as described herein, at least one segment of which is complementary to an mRNA within the cells of the hemipteran pest. A dsRNA molecule, including its modified form such as an siRNA, miRNA, shRNA, or hpRNA molecule, ingested by a hemipteran pest in accordance with the invention may be at least from about 80%, 81%, 82%, 83%, 84%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or about 100% identical to an RNA molecule transcribed from a chromatin remodeling gene DNA molecule, for example, comprising a polynucleotide selected from the group consisting of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, and SEQ ID NO:67. Isolated and substantially purified nucleic acid molecules including, but not limited to, non-naturally occurring polynucleotides and recombinant DNA constructs for providing dsRNA molecules of the present invention are therefore provided, which suppress or inhibit the expression of an endogenous coding polynucleotide or a target coding polynucleotide in the hemipteran pest when introduced thereto.

Particular embodiments provide a delivery system for the delivery of iRNA molecules for the post-transcriptional inhibition of one or more target gene(s) in a hemipteran plant pest and control of a population of the plant pest. In some embodiments, the delivery system comprises ingestion of a host transgenic plant cell or contents of the host cell comprising RNA molecules transcribed in the host cell. In these and further embodiments, a transgenic plant cell or a transgenic plant is created that contains a recombinant DNA construct providing a stabilized dsRNA molecule of the invention. Transgenic plant cells and transgenic plants comprising nucleic acids encoding a particular iRNA molecule may be produced by employing recombinant DNA technologies (which basic technologies are well-known in the art) to

construct a plant transformation vector comprising a polynucleotide encoding an iRNA molecule of the invention (e.g., a stabilized dsRNA molecule); to transform a plant cell or plant; and to generate the transgenic plant cell or the transgenic plant that contains the transcribed iRNA molecule.

To impart protection from hemipteran pests to a transgenic plant, a recombinant DNA molecule may, for example, be transcribed into an iRNA molecule, such as a dsRNA molecule, an siRNA molecule, a miRNA molecule, a shRNA molecule, or a hpRNA molecule. In some embodiments, an RNA molecule transcribed from a recombinant DNA molecule may form a dsRNA molecule within the tissues or fluids of the recombinant plant. Such a dsRNA molecule may be comprised in part of a polynucleotide that is identical to a corresponding polynucleotide transcribed from a DNA within a hemipteran pest of a type that may infest the host plant. Expression of a target gene within the hemipteran pest is suppressed by the dsRNA molecule, and the suppression of expression of the target gene in the hemipteran pest results in the transgenic plant being resistant to the pest. The modulatory effects of dsRNA molecules have been shown to be applicable to a variety of genes expressed in pests, including, for example, endogenous genes responsible for cell division, chromosomal remodeling, and cellular metabolism or cellular transformation, including housekeeping genes; transcription factors; molting-related genes; and other genes which encode polypeptides involved in cellular metabolism or normal growth and development.

For transcription from a transgene in vivo or an expression construct, a regulatory region (e.g., promoter, enhancer, silencer, and polyadenylation signal) may be used in some embodiments to transcribe the RNA strand (or strands). Therefore, in some embodiments, as set forth, supra, a polynucleotide for use in producing iRNA molecules may be operably linked to one or more promoter elements functional in a plant host cell. The promoter may be an endogenous promoter, normally resident in the host genome. The polynucleotide of the present invention, under the control of an operably linked promoter element, may further be flanked by additional elements that advantageously affect its transcription and/or the stability of a resulting transcript. Such elements may be located upstream of the operably linked promoter, downstream of the 3' end of the expression construct, and may occur both upstream of the promoter and downstream of the 3' end of the expression construct.

In embodiments, suppression of a target gene (e.g., a chromatin remodeling gene) results in a parental RNAi phenotype; a phenotype that is observable in progeny of the subject (e.g., a hemipteran pest) contacted with the iRNA molecule. In some embodiments, the pRNAi phenotype comprises the pest being rendered less able to produce viable offspring. In particular examples of pRNAi, a nucleic acid that initiates pRNAi does not increase the incidence of mortality in a population into which the nucleic acid is delivered. In other examples of pRNAi, a nucleic acid that initiates pRNAi also increases the incidence of mortality in a population into which the nucleic acid is delivered.

In some embodiments, a population of hemipteran pests is contacted with an iRNA molecule, thereby resulting in pRNAi, wherein the pests survive and mate but produce eggs that are less able to hatch viable progeny than eggs produced by pests of the same species that are not provided the nucleic acid(s). In some examples, such pests do not lay eggs or lay fewer eggs than what is observable in pests of the same species that are not contacted with the iRNA molecule.

In some examples, the eggs oviposited by such pests do not hatch or hatch at a rate that is significantly less than what is observable in pests of the same species that are not contacted with the iRNA molecule. In some examples, the nymphs that hatch from eggs oviposited by such pests are not viable or are less viable than what is observable in pests of the same species that are not contacted with the iRNA molecule.

Transgenic crops that produce substances that provide protection from insect feeding are vulnerable to adaptation by the target insect pest population reducing the durability of the benefits of the insect protection substance(s). Traditionally, delays in insect pest adaptation to transgenic crops are achieved by (1) the planting of "refuges" (crops that do not contain the pesticidal substances, and therefore allow survival of insects that are susceptible to the pesticidal substance(s)); and/or (2) combining insecticidal substances with multiple modes of action against the target pests, so that individuals that are resistant to one mode of action are killed by a second mode of action.

In some examples, iRNA molecules (e.g., expressed from a transgene in a host plant) represent new modes of action for combining with *Bacillus thuringiensis* insecticidal protein technology (e.g., Cry1A, Cry2A, Cry3A, Cry11A, and Cry51A) and/or lethal RNAi technology in Insect Resistance Management gene pyramids to mitigate against the development of insect populations resistant to either of these control technologies.

Parental RNAi may result in some embodiments in a type of pest control that is different from the control obtained by lethal RNAi, and which may be combined with lethal RNAi to result in synergistic pest control. Thus, in particular embodiments, iRNA molecules for the post-transcriptional inhibition of one or more target gene(s) in a hemipteran plant pest can be combined with other iRNA molecules to provide redundant RNAi targeting and synergistic RNAi effects.

Parental RNAi (pRNAi) that causes egg mortality or loss of egg viability has the potential to bring further durability benefits to transgenic crops that use RNAi and other mechanisms for insect protection. pRNAi prevents exposed insects from producing progeny, and therefore from passing on to the next generation any alleles they carry that confer resistance to the pesticidal substance(s). pRNAi is particularly useful in extending the durability of insect-protected transgenic crops when it is combined with one or more additional pesticidal substances that provide protection from the same pest populations. Such additional pesticidal substances may in some embodiments include, for example, nymph-active dsRNA; insecticidal proteins (such as those derived from *Bacillus thuringiensis*, *Alcaligenes* spp., *Pseudomonas* spp., or other organisms); and other insecticidal substances. This benefit arises because insects that are resistant to the pesticidal substances occur as a higher proportion of the population in the transgenic crop than in the refuge crop. If a ratio of resistance alleles to susceptible alleles that are passed on to the next generation is lower in the presence of pRNAi than in the absence of pRNAi, the evolution of resistance will be delayed.

For example, pRNAi may not reduce the number of individuals in a first pest generation that are inflicting damage on a plant expressing an iRNA molecule. However, the ability of such pests to sustain an infestation through subsequent generations may be reduced. Conversely, lethal RNAi may kill pests that already are infesting the plant. When pRNAi is combined with lethal RNAi, pests that are contacted with a parental iRNA molecule may breed with pests from outside the system that have not been contacted with the iRNA, however, the progeny of such a mating may

be non-viable or less viable, and thus may be unable to infest the plant. At the same time, pests that are contacted with a lethal iRNA molecule may be directly affected. The combination of these two effects may be synergistic; i.e., the combined pRNAi and lethal RNAi effect may be greater than the sum of the pRNAi and lethal RNAi effects independently. pRNAi may be combined with lethal RNAi, for example, by providing a plant that expresses both lethal and parental iRNA molecules; by providing in the same location a first plant that expresses lethal iRNA molecules and a second plant that expresses parental iRNA molecules; and/or by contacting female and/or male pests with the pRNAi molecule, and subsequently releasing the contacted pests into the plant environment, such that they can mate unproductively with the plant pests.

Some embodiments provide methods for reducing the damage to a host plant (e.g., a soybean plant) caused by a hemipteran pest that feeds on the plant, wherein the method comprises providing in the host plant a transformed plant cell expressing at least one nucleic acid molecule of the invention, wherein the nucleic acid molecule(s) functions upon being taken up by the pest(s) to inhibit the expression of a target polynucleotide within the pest(s), which inhibition of expression results in reduced reproduction, for example, in addition to mortality and/or reduced growth of the pest(s), thereby reducing the damage to the host plant caused by the pest. In some embodiments, the nucleic acid molecule(s) comprise dsRNA molecules. In these and further embodiments, the nucleic acid molecule(s) comprise dsRNA molecules that each comprise more than one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell. In some embodiments, the nucleic acid molecule(s) consist of one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell.

In some embodiments, a method for increasing the yield of a corn crop is provided, wherein the method comprises introducing into a corn plant at least one nucleic acid molecule of the invention; and cultivating the corn plant to allow the expression of an iRNA molecule comprising the nucleic acid, wherein expression of an iRNA molecule comprising the nucleic acid inhibits hemipteran pest damage and/or growth, thereby reducing or eliminating a loss of yield due to hemipteran pest infestation. In some embodiments, the iRNA molecule is a dsRNA molecule. In these and further embodiments, the nucleic acid molecule(s) comprise dsRNA molecules that each comprise more than one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell. In some embodiments, the nucleic acid molecule(s) consists of one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell.

In some embodiments, a method for increasing the yield of a plant crop is provided, wherein the method comprises introducing into a female hemipteran pest (e.g., by injection, by ingestion, by spraying, and by expression from a DNA) at least one nucleic acid molecule of the invention; and releasing the female pest into the crop, wherein mating pairs including the female pest are unable or less able to produce viable offspring, thereby reducing or eliminating a loss of yield due to hemipteran pest infestation. In particular embodiments, such a method provides control of subsequent generations of the pest. In similar embodiments, the method comprises introducing the nucleic acid molecule of the invention into a male hemipteran pest, and releasing the male pest into the crop (e.g., wherein pRNAi male pests produce less sperm than untreated controls). In some

embodiments, the nucleic acid molecule is a DNA molecule that is expressed to produce an iRNA molecule. In some embodiments, the nucleic acid molecule is a dsRNA molecule. In these and further embodiments, the nucleic acid molecule(s) comprise dsRNA molecules that each comprise more than one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell. In some embodiments, the nucleic acid molecule(s) consists of one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell.

In some embodiments, a method for modulating the expression of a target gene in a hemipteran pest is provided, the method comprising: transforming a plant cell with a vector comprising a polynucleotide encoding at least one iRNA molecule of the invention, wherein the polynucleotide is operatively-linked to a promoter and a transcription termination element; culturing the transformed plant cell under conditions sufficient to allow for development of a plant cell culture including a plurality of transformed plant cells; selecting for transformed plant cells that have integrated the polynucleotide into their genomes; screening the transformed plant cells for expression of an iRNA molecule encoded by the integrated polynucleotide; selecting a transgenic plant cell that expresses the iRNA molecule; and feeding the selected transgenic plant cell to the hemipteran pest. Plants may also be regenerated from transformed plant cells that express an iRNA molecule encoded by the integrated nucleic acid molecule. In some embodiments, the iRNA molecule is a dsRNA molecule. In these and further embodiments, the nucleic acid molecule(s) comprise dsRNA molecules that each comprise more than one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell. In some embodiments, the nucleic acid molecule(s) consists of one polynucleotide that is specifically hybridizable to a nucleic acid molecule expressed in a hemipteran pest cell.

iRNA molecules of the invention can be incorporated within the seeds of a plant species (e.g., soybean), either as a product of expression from a recombinant gene incorporated into a genome of the plant cells, or as incorporated into a coating or seed treatment that is applied to the seed before planting. A plant cell comprising a recombinant gene is considered to be a transgenic event. Also included in embodiments of the invention are delivery systems for the delivery of iRNA molecules to hemipteran pests. For example, the iRNA molecules of the invention may be directly introduced into the cells of a pest(s). Methods for introduction may include direct mixing of iRNA into the diet of the hemipteran pest (e.g., by mixing with plant tissue from a host for the pest), as well as application of compositions comprising iRNA molecules of the invention to host plant tissue. For example, iRNA molecules may be sprayed onto a plant surface. Alternatively, an iRNA molecule may be expressed by a microorganism, and the microorganism may be applied onto the plant surface, or introduced into a root or stem by a physical means such as an injection. As discussed, supra, a transgenic plant may also be genetically engineered to express at least one iRNA molecule in an amount sufficient to kill the hemipteran pests known to infest the plant. iRNA molecules produced by chemical or enzymatic synthesis may also be formulated in a manner consistent with common agricultural practices, and used as spray-on or bait products for controlling plant damage by a hemipteran pest. The formulations may include the appropriate adjuvants (e.g., stickers and wetters) required for efficient foliar coverage, as well as UV protectants to protect

iRNA molecules (e.g., dsRNA molecules) from UV damage. Such additives are commonly used in the bioinsecticide industry, and are well known to those skilled in the art. Such applications may be combined with other spray-on insecticide applications (biologically based or otherwise) to enhance plant protection from hemipteran pests.

All references, including publications, patents, and patent applications, cited herein are hereby incorporated by reference to the extent they are not inconsistent with the explicit details of this disclosure, and are so incorporated to the same extent as if each reference were individually and specifically indicated to be incorporated by reference and were set forth in its entirety herein. The references discussed herein are provided solely for their disclosure prior to the filing date of the present application. Nothing herein is to be construed as an admission that the inventors are not entitled to antedate such disclosure by virtue of prior invention.

The following Examples are provided to illustrate certain particular features and/or aspects. These Examples should not be construed to limit the disclosure to the particular features or aspects described.

EXAMPLES

Example 1: Identification of Candidate Target Genes

RNAi Target Selection.

In one example, six stages of BSB development were selected for mRNA library preparation. Additional samples were prepared using BSB midgut and salivary glands. Brown stink bug midguts and salivary glands were dissected from 10 and 25 mixed sex adults respectively under a dissecting microscope on a chilled clean glass slide and immediately frozen on dry ice. Total RNA was extracted from insects frozen at -70° C. and homogenized in 10 volumes of Lysis/Binding buffer in Lysing MATRIX A 2 mL tubes (MP BIOMEDICALS, Santa Ana, Calif.) on a Fast-Prep®-24 Instrument (MP BIOMEDICALS). Total mRNA was extracted using a mirVana™ miRNA Isolation Kit (AMBION; INVITROGEN) according to the manufacturer's protocol. RNA sequencing using an Illumina® HiSeq™ system (San Diego, Calif.) provided candidate target gene sequences for use in RNAi insect control technology. HiSeq™ generated a total of about 378 million reads for the six samples. The reads were assembled individually for each sample using TRINITY assembler software (Grabherr et al. (2011) Nature Biotech. 29:644-652). The assembled transcripts were combined to generate a pooled transcriptome. This BSB pooled transcriptome contains 378,457 sequences.

BSB_Brahma, mi-2, iswi-1, iswi-2, chd1, ino80, and Domino Ortholog Identification.

tBLASTn searches of the BSB pooled transcriptome were performed using sequences of the *Drosophila* BRAHMA (brm-PA, GENBANK Accession No. NP_536745 and NP_536746), MI-2 (Mi-2-PA, GENBANK Accession No. NP_001014591.1, NP_001163476.1, NP_001262078.1, NP_649154.2, and NP_001014591.1), ISWI (Iswi-PA, GENBANK Accession No. NP_523719, NP_725203, and NP_725204), and CHD1 (Chd1-PA, GENBANK Accession No. NP_477197 and NP_001245851) proteins as queries. BSB brahma (SEQ ID NO:1; SEQ ID NO:63), mi-2 (SEQ ID NO:8; SEQ ID NO:64), iswi-1 (SEQ ID NO:10; SEQ ID NO:65), iswi-2 (SEQ ID NO:12; SEQ ID NO:66), chd1 (SEQ ID NO:14; SEQ ID NO:67), ino80 (SEQ ID NO:30), and domino (SEQ ID NO:32) transcripts were identified as BSB candidate target genes.

BSB homology info. The BSB brahma (SEQ ID NO:1) is somewhat (72% identity) related to a fragment of a sequence from *Ciona intestinalis* (GENBANK Accession No. AK116913.1). The closest homolog of the BSB BRAHMA amino acid sequence (SEQ ID NO:2) is a *Camponotus floridanus* protein having GENBANK Accession No. EFN67856.1 (79% similar; 70% identical over the homology region). The BSB mi-2 (SEQ ID NO:8) is somewhat (76% identity) related to a fragment of a sequence from *Acyrtosiphon pisum* (GENBANK Accession No. XM_008186702.1). The closest homolog of the BSB MI-2 amino acid sequence (SEQ ID NO:9) is a *Bombus impatiens* protein having GENBANK Accession No. XP_003493868.1 (79% similar; 71% identical over the homology region). The BSB iswi-1 (SEQ ID NO:10) is somewhat (75% identity) related to a fragment of a sequence from *Bombus impatiens* (GENBANK Accession No. XM_003486758.1). The closest homologs of the BSB ISWI-1 amino acid sequence (SEQ ID NO:11) are a *Megachile rotundata* and *Apis dorsata* proteins having GENBANK Accession Nos. XP_003708682.1 and XP_006615660.1 respectively (91% similar; 84% identical over the homology region). The BSB iswi-2 (SEQ ID NO:12) is somewhat (76% identity) related to a fragment of a sequence from *Latimeria chalumnae* (GENBANK Accession No. XM_005994941.1). The closest homolog of the BSB ISWI-2 amino acid sequence (SEQ ID NO:13) is a *Cerapachys biroi* protein having GENBANK Accession No. EZA60706.1 (93% similar; 84% identical over the homology region). The BSB chd1 (SEQ ID NO:14) is somewhat (77% identity) related to a fragment of a sequence from *Apis mellifera* (GENBANK Accession No. XM_006565933.1). The closest homolog of the BSB CHD1 amino acid sequence (SEQ ID NO:15) is a *Riptortus pedestris* protein having GENBANK Accession No. BAN20905.1 (94% similar; 88% identical over the homology region). The BSB ino80 (SEQ ID NO:30) is somewhat (79% identity) related to a fragment of a sequence from *Hydra magnipapillata* (GENBANK Accession No. XM_002164516.2). The closest homolog of the BSB INO80 amino acid sequence (SEQ ID NO:31) is a *Zootermopsis nevadensis* protein having GENBANK Accession No. KDR11347.1 (74% similar; 64% identical over the homology region). The BSB domino (SEQ ID NO:32) is somewhat (80% identity) related to a fragment of a sequence from *Apis florea* (GENBANK Accession No. XR_143356.1). The closest homolog of the BSB DOMINO amino acid sequence (SEQ ID NO:33) is a *Nasonia vitripennis* protein having GENBANK Accession No. XP_008210745.1 (71% similar; 58% identical over the homology region).

These genes encode SNF2-type chromatin remodeler proteins, which correspond to a subunit of the chromatin remodeler complexes that play global roles in mobilizing nucleosomes. See, for example, Brizuela et al. (supra); Kal et al. (2000) Genes Devel. 14:1058-71; and Tamkun et al. (1992) Cell 68:561-72. Although they share a SNF2-Helicase domain, most chromatin remodelers within each species have non-redundant functions that are conferred by the additional domains they comprise. These characteristics present chromatin remodeling ATPases as attractive targets for multi-generational/parental RNAi.

The SWI2/SNF2 (mating type switch/sucrose non-fermenting) family of the ATP-dependent remodeling enzymes contains a bromodomain, which binds acetylated histones. While yeasts and vertebrates contain several SWI2/SNF2 proteins, only one SWI2/SNF2 protein, BRAHMA, has been identified in *Drosophila*. BRAHMA is well-conserved, and yet distinct, from other insect SNF2-containing proteins,

with the putative orthologs clustering closely on a phylogenetic tree. FIG. 2. The human BRAHMA (BRM) as well as the *Saccharomyces cerevisiae* SNF2 protein cluster together with insect BRAHMAs. Furthermore, the orthologs of the *Drosophila* BRAHMA maintain overall protein domain conservation including the SNF2 ATPase/helicase, the bromodomain as well as additional domains: conserved Gln, Leu, Gln motif domain (QLQ), DNA-binding HSA domain, and BRK (brahma and kismet) domain. FIG. 3A.

BRAHMA is known to incorporate into BAP (Brahma Associated Proteins) and PBAP (Polybromo-associated BAP) chromatin remodeling complexes. The loss of *Drosophila* brahma impairs overall transcription by RNA polymerase II (Pol II), suggesting a broad function for the BRAHMA complexes. In *Drosophila*, the maternal contribution of brahma is needed for early embryogenesis, while the zygotic brahma expression is necessary for late embryonic development. In addition to embryogenesis, *Drosophila* brahma is involved in gametogenesis. Brahma RNAi-treated female BSB produce no viable eggs. Table 5. Further, BSB females whose brahma was depleted via RNAi lay no eggs altogether. Tables 3 and 4.

The ISWI (Imitation SWI/imitation switch) family is defined by histone-binding domain that comprises the HAND, SANT, and SLIDE domains in a HAND-SANT-SLIDE architecture (also annotated as HAND-SLIDE). In *Drosophila*, the ISWI family of ATP-dependent remodeling enzymes has only one member, ISWI. The *Drosophila* ISWI can confer multiple functions by integrating into various complexes that include ATP-dependent chromatin assembly and remodeling factor (ACF), nucleosome remodeling factor (NURF), and chromatin accessibility complex (CHRAC). Loss of ISWI in *Drosophila* results in dramatic chromosome condensation defects.

BSB express at least two iswi homologs (SEQ ID NO:10 and SEQ ID NO:12 (with SEQ ID NO:12 being partial sequence). The complete BSB ISWI protein contains the SNF2 ATPase/helicase, HAND-SANT-SLIDE (identified as HAND and SLIDE by Pfam) and DNA-binding domain (DBINO). FIG. 3B. The identified ISWI-2 protein from BSB comprises only HAND-SANT-SLIDE domains. FIG. 3B. The contig that comprises iswi-2 (SEQ ID NO:12) is 1316 nucleotides long; based on the alignment with known *Drosophila* ISWI protein this contig does not contain the first half of the ISWI protein sequence. Therefore, it is reasonable to assume that the current BSB transcriptome assembly contains an incomplete sequence of iswi-2 transcript.

The parental RNAi applications of both BSB_iswi-1 and BSB_iswi-2 result in both egg laying and egg hatch defects. Tables 4 and 5.

Proteins of the CHD (chromodomain helicase DNA-binding) family of ATP-dependent remodeling enzymes contain two amino-terminal chromodomains [chromatin organization modifier]. FIG. 3C. The *Drosophila* CHD proteins include CHD1, MI-2, CHD3, and KISMET. The CHD family is further subdivided into three subfamilies, herein referred to as subfamilies I, II, and III. The *Drosophila* CHD1 belongs to CHD subfamily I, which has a C-terminal DNA-binding domain. FIG. 1C (DUF4208). In *Drosophila*, CHD1 protein shows similar distribution patterns to BRAHMA, yet chd1 mutant flies are viable. Interestingly, the *Drosophila* chd1 is needed for gametogenesis. BSB females subjected to chd1 RNAi show a significant decrease in both egg production and hatch rates. Tables 4 and 5.

MI-2 and CHD3 belong to subfamily II. Enzymes of the CHD subfamily II have no DNA-binding domain, but have Zn-finger-like domains called PHD (plant homeodomain)

fingers. The BSB ortholog of MI-2 mirrors the *Drosophila* domain arrangement, and includes the SNF2 ATPase/helicase domain, the double chromodomain, PHD fingers, and CHDNT domain that is associated with PHD finger-containing chromodomain helicases, as well as other conserved domains of unknown functions, DUF1087 and DUF1086. FIG. 3D. The *Drosophila* MI-2 is known to associate with the NuRD (Nucleosome Remodeling Deacetylase) and dMec (*Drosophila* MEP-1 containing complex) complexes. Maternal expression of mi-2 is necessary for gametogenesis. BSB females whose mi-2 was depleted via RNAi lay very few eggs. Table 4.

The third subfamily of CHD proteins is represented by KISMET in *Drosophila*; in humans this subfamily comprises CHD5-9. Like other CHD proteins, KISMET contains an SNF2 domain and a chromodomain. FIG. 3E. Unlike other CHD subfamilies, KISMET has characteristics of both CHD and SWI2/SNF2 proteins, in that it has a BRK domain that is common to both BRAHMA and KISMET. Although BRK is a well-established feature of *Drosophila* KISMET, a standard Pfam analysis did not identify this domain in BSB. FIG. 3E. Loss of either maternal or zygotic function of kismet causes defects during *Drosophila* embryogenesis and the insects die during early larval stages, while oogenesis is unaffected.

Example 2: Degenerate Sequences Comprising Chromatin Remodelers

Brahma and its homologs, as well as mi-2 and other chromatin remodelers and their orthologs, share the same functional domains and sequence-level conservation. RNAi target sites were designed within the conserved SNF2 family N-terminal and Helicase C-terminal domains (here referred to as SNF2-Helicase) that are common to all chromatin remodelers, as well as chromatin binding and other functional domains that are conserved within each family (including bromodomain, chromodomain, and HAND-SLIDE domains). RNAi target sequences that are common to *Diabrotica virgifera virgifera*, *Euschistus heros*, *Tribolium castaneum*, and *Drosophila melanogaster* were designed. The DNA nucleotides and RNAi nucleotides are listed according to the standard IUPAC code:

A=Adenine
C=Cytosine
G=Guanine
T=Thymine
R=A or G
Y=C or T
S=G or C
W=A or T
K=G or T
M=A or C
B=C or G or T
D=A or G or T
H=A or C or T
V=A or C or G
N=A or C or G or T

dsRNA encoding sequences targeting SNF2-Helicase regions (SEQ ID NOs:34-37) and chromatin remodeling domains (SEQ ID NOs:38-41) were designed by aligning the amino acid sequences for each target protein from four species, *Diabrotica v. virgifera*, *E. heros*, *Tribolium castaneum*, and *Drosophila melanogaster*, using Vector NTI Align X (Invitrogen, Grand Island, N.Y.). Highly homologous regions of the amino acid sequence containing at least 8 amino acids within the SNF2 domain or chromatin remodel-

eling domain specific to each target protein were selected. The corresponding nucleotide sequence for each species from each target was then aligned also using the Align X program. Where there was a misalignment across the four species the nucleotides were replaced with nucleotides as shown above. Finally, the sequence was aligned against the nucleotide sequence from *Apis mellifera* to determine if the sequence would also target that species. If the sequence could also target the protein from *A. mellifera* either new regions were chosen or the sequence was shortened to at least 21 bases which did not target *A. mellifera* proteins.

Example 3: Preparation of RNAi Molecules

Template Preparation and dsRNA Synthesis.

cDNA was prepared from total BSB RNA extracted from a single young adult insect (about 90 mg) using TRIzol® Reagent (LIFE TECHNOLOGIES, Grand Island, N.Y.). The insect was homogenized at room temperature in a 1.5 mL microcentrifuge tube with 200 µL of TRIzol® using a pellet pestle (FISHERBRAND, Grand Island, N.Y.) and Pestle Motor Mixer (COLE-PARMER, Vernon Hills, Ill.). Following homogenization, an additional 800 µL of TRIzol® was added, the homogenate was vortexed, and then incubated at room temperature for five minutes. Cell debris was removed by centrifugation and the supernatant was transferred to a new tube. Following manufacturer-recommended TRIzol® extraction protocol for 1 mL of TRIzol®, the RNA pellet

using a NANODROP™ 8000 spectrophotometer (THERMO SCIENTIFIC, Wilmington, Del.).

cDNA was reverse-transcribed from 5 µg BSB total RNA template and oligo dT primer using a SUPERScript III FIRST-STRAND SYNTHESIS SYSTEM™ for RT-PCR (INVITROGEN), following the supplier's recommended protocol. The final volume of the transcription reaction was brought to 100 µL with nuclease-free water.

Primers were used to amplify DNA templates for dsRNA transcription. Table 1. The DNA templates were amplified using "touch-down" PCR (annealing temperature lowered from 60° C. to 50° C. in a 1° C./cycle decrease) with 1 µL cDNA (above) as the template. Fragments comprising a 499 bp segment of brahma (i.e., BSB_brm-1; SEQ ID NO:3), a 496 bp segment of mi-2 (i.e., BSB_mi-2-1; SEQ ID NO:16), a 481 bp segment of iswi-1 (i.e., BSB_iswi-1-1; SEQ ID NO:17), a 490 bp segment of iswi-2 (i.e., BSB_iswi-2-1; SEQ ID NO:18), and a 496 bp segment of chd1 (i.e., BSB_chd1-1; SEQ ID NO:19) were generated during 35 cycles of PCR. A 301 pb template for dsRNA termed YFPv2 (SEQ ID NO:5) was synthesized using primers YFPv2_F (SEQ ID NO:6) and YFPv2_R (SEQ ID NO:7). The BSB-specific and YFPv2 primers contained a T7 phage promoter sequence (SEQ ID NO:4) at their 5' ends, enabling the use of the aforementioned BSB DNA fragments for dsRNA transcription.

TABLE 1

Primer pairs used to amplify DNA templates for dsRNA transcription.			
	Gene (Region)	Primer_ID	Sequence
Pair 1	Brahma	BSB_brm-1-F	TTAATACGACTCACTATAGGAGAGATGATGAAGAAGATGCAAGTAC (SEQ ID NO: 20)
		BSB_brm-1-R	TTAATACGACTCACTATAGGAGACTCCACTCCCTCGGGTC (SEQ ID NO: 21)
Pair 2	mi-2	BSB_Mi-2-1-F	TTAATACGACTCACTATAGGAGAGACTACCTCGAGGGTGAAGG (SEQ ID NO: 22)
		BSB_Mi-2-1-R	TTAATACGACTCACTATAGGAGAGTAATCTCTCAACAGCTTTATCGTC (SEQ ID NO: 23)
Pair 3	iswi-1	BSB_Iswi-1-1-F	TTAATACGACTCACTATAGGAGACAAAAAT TGAACTGACCGTTCTAG (SEQ ID NO: 24)
		BSB_Iswi-1-1-R	TTAATACGACTCACTATAGGAGAGCTAATGTTGATTTTGGTACGATG (SEQ ID NO: 25)
Pair 4	iswi-2	BSB_Iswi-2-1-F	TTAATACGACTCACTATAGGAGAGTTCAAGATTTCCAATTTTCCAC (SEQ ID NO: 26)
		BSB_Iswi-2-1-R	TTAATACGACTCACTATAGGAGAGAAACGGTGCTCTATATCGACTC (SEQ ID NO: 27)
Pair 5	chd1	BSB_Chdl-1-F	TTAATACGACTCACTATAGGAGACAGCTGGAACCATATATTCTACGAC (SEQ ID NO: 28)
		BSB_Chdl-1-R	TTAATACGACTCACTATAGGAGAGTGAATTTTCAGCATTGAAATGATCG (SEQ ID NO: 29)
Pair 6	YFPv2	YFPv2_F	TTAATACGACTCACTATAGGAGAGCATCTGGAGCACTTCTCTTCA (SEQ ID NO: 6)
		YFPv2_R	TTAATACGACTCACTATAGGAGACCATCTCCTTCAAGGTGATTG (SEQ ID NO: 7)

was dried at room temperature and resuspended in 200 µL Tris Buffer from a GFX PCR DNA AND GEL EXTRACTION KIT (Illustra™; GE HEALTHCARE LIFE SCIENCES, Pittsburgh, Pa.) using Elution Buffer Type 4 (i.e. 10 mM Tris-HCl pH8.0). RNA concentration was determined

dsRNAs were synthesized using 2 µL of PCR product (above) as the template with a MEGAscript™ RNAi kit (AMBION) or HiScribe® T7 In Vitro Transcription Kit, used according to the manufacturer's instructions. See FIG. 1B. dsRNA was quantified on a NANODROP™ 8000

spectrophotometer and diluted to 1 µg/µL in nuclease-free 0.1×TE buffer (1 mM Tris HCL, 0.1 mM EDTA, pH 7.4).

Example 4: Brahma dsRNA Injection of 2nd Instar *Euschistus heros* Nymphs

Insect Rearing.

Neotropical Brown Stink Bugs (BSB; *Euschistus heros*) were reared in a 27° C. incubator, at 65% relative humidity, with 16:8 hour light:dark cycle. One gram of eggs collected over 2-3 days was seeded in 5 L containers with filter paper discs at the bottom; the containers were covered with #18 mesh for ventilation. Each rearing container yielded approximately 300-400 adult BSB. At all stages, the insects were fed fresh green beans three times per week and a sachet of seed mixture containing sunflower seeds, soybeans, and peanuts (3:1:1 by weight ratio) was replaced once a week. Water was supplemented in vials with cotton plugs as wicks. After the initial two weeks, insects were transferred to a new container once a week.

BSB Artificial Diet.

BSB artificial diet was prepared as follows and used within two weeks of preparation. Lyophilized green beans were blended to a fine powder in a MAGIC BULLET® blender while raw (organic) peanuts were blended in a separate MAGIC BULLET® blender. Blended dry ingredients were combined (weight percentages: green beans, 35%; peanuts, 35%; sucrose, 5%; Vitamin complex (e.g. Vanderzant Vitamin Mixture for insects, SIGMA-ALDRICH), 0.9%); in a large MAGIC BULLET® blender, which was capped and shaken well to mix the ingredients. The mixed dry ingredients were then added to a mixing bowl. In a separate container, water and benomyl anti-fungal agent (50 ppm; 25 µL 20,000 ppm solution/50 mL diet solution) were mixed well and then added to the dry ingredient mixture. All ingredients were mixed by hand until the solution was fully blended. The diet was shaped into desired sizes, wrapped loosely in aluminum foil, heated for 4 hours at 60° C., then cooled and stored at 4° C.

Injection of dsRNA into BSB Hemocoel.

BSB were reared on a green bean and seed diet, as the colony described above, in a 27° C. incubator at 65% relative humidity and 16:8 hour light:dark photoperiod. Second instar nymphs (each weighing 1 to 1.5 mg) were gently handled with a small brush to prevent injury and were placed in a Petri dish on ice to chill and immobilize the insects. Each insect was injected with 55.2 nL of a 500 ng/µL dsRNA solution (i.e., 27.6 ng dsRNA; dosage of 18.4 to 27.6 µg/g body weight). Injections were performed using a NANOJECT™ II injector (DRUMMOND SCIENTIFIC, Broomhall, Pa.) equipped with an injection needle pulled from a Drummond 3.5 inch #3-000-203-G/X glass capillary. The needle tip was broken and the capillary was backfilled with light mineral oil, then filled with 2 to 3 µL dsRNA. dsRNA was injected into the abdomen of the nymphs (10 insects injected per dsRNA per trial), and the trials were repeated on three different days. Injected insects (5 per well) were transferred into 32-well trays (Bio-RT-32 Rearing Tray; BIO-SERV, Frenchtown, N.J.) containing a pellet of artificial BSB diet and covered with Pull-N-Peel™ tabs (BIO-CV-4; BIO-SERV). Moisture was supplied by means of 1.25 mL water in a 1.5 mL microcentrifuge tube with a cotton wick. The trays were incubated at 26.5° C., 60% humidity and 16:8 hour light:dark photoperiod. Viability counts and weights were taken on day 7 after the injections.

Injection of dsRNA that Targets Brahma mRNA in BSB 2nd Instar Nymphs.

dsRNA that targets segment of YFP coding region, YFPv2, was used as a negative control in BSB injection experiments. As summarized in Table 2, at least ten 2nd instar BSB nymphs (1-1.5 mg each) were injected into the hemocoel with 55.2 nL BSB_brm-1 (500 ng/µL) for an approximate final concentration of 18.4-27.6 µg dsRNA/g insect. Percent mortality was scored seven days after dsRNA injection. The mortality determined for BSB_brm-1 dsRNA was not significantly different from that seen with the same amount of injected YFPv2 dsRNA (negative control), with p=0.279 (Student's t-test). There was also no significant difference between the YFPv2 dsRNA injected and not injected treatments.

TABLE 2

Results of BSB_brm-1 dsRNA injection into the hemocoel of 2 nd instar <i>E. heros</i> nymphs seven days after injection. Table shows mean percent mortality, N number of trials, and standard error of the mean (SEM). Means comparisons were performed with YFP dsRNA as control, using a Student's t-test with Dunnett's adjustment in JMP ® Pro 11; p-value shown.				
Treatment	Mean % mortality	SEM	N trials	t-test (p)
BSB_brm-1	27	12.0	3	0.3039
not injected	13	3.3	3	0.9384
YFPv2 dsRNA	10	5.8	3	

*Ten insects injected per trial for each dsRNA.

Example 5: Parental RNAi Effects Following dsRNA Injection in *Euschistus heros*

Injection of dsRNA into BSB Hemocoel.

BSB were reared as described above for the colony. In the following exemplification, young adults (up to one week post adult molt) were collected and chilled in a secondary container on ice. The females and males were separated based on structural dimorphism of the genitalia. Female BSB were handled with Featherweight entomology forceps and injected with dsRNA using a NANOJECT™ II injector (DRUMMOND SCIENTIFIC, Broomhall, Pa.) equipped with an injection needle pulled from a Drummond 3.5 inch #3-000-203-G/X glass capillary. The needle tip was broken and the capillary was backfilled with light mineral oil then filled with 3 µL dsRNA. Ten to twenty females (approximately 90 mg each) per treatment were injected with dsRNA. Each female was injected into the abdomen twice consecutively with 69 nL 1 µg/µL dsRNA for a total of 138 nL (138 ng). Each batch of ten females was moved into a 1 quart (~950 mL) bin with an opening in the lid and #18 mesh for ventilation. Two adult males were added to each bin of ten females. The insects were supplied with a vial of water, green beans, and seeds as described in the rearing procedure. The insects were kept at 26.5° C., 60% humidity and 16:8 light:dark photoperiod.

Surviving female counts, oviposition, and egg hatch numbers were collected on a daily basis starting seven to nine days after injection and continued for up to 16 days. Eggs were removed daily and kept in Petri dishes or multi-well plates on a layer of 1% agarose in water. The adult insects were transferred into bins with fresh water and food every week.

Injections of dsRNA that target brahma, iswi-1, iswi-2, mi-2, and/or chd1 in BSB females decreased egg laying. Females injected with dsRNA that targets a 301 nt sequence of the YFP coding region were used as a negative controls, and compared to un-injected and females injected with

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BSB_brm-1 dsRNA (SEQ ID NO:3). As summarized in Table 3, un-injected females did not lay statistically different numbers of eggs from YFPv2 controls. On the other hand, BSB_brm-1 dsRNA-injected females oviposited no eggs.

Injection of 138 ng chromatin-remodeling ATPase dsRNA had no effect on viability or no immediate effect on viability of the adult female BSB. FIG. 4A. Injection of BSB brahma dsRNA (BSB_brm-1 (SEQ ID NO:3)) and of dsRNAs that target BSB_mi-2-1 (SEQ ID NO:16), BSB_iswi-1-1 (SEQ ID NO:17), BSB_iswi-2-1 (SEQ ID NO:18), and BSB_chd1-1 (SEQ ID NO:19) of BSB greatly decreased oviposition or eliminated oviposition altogether, as compared to negative YFPv2 dsRNA controls (SEQ ID NO:5) Table 4 and FIG. 4B. Oviposition by BSB females injected with dsRNAs BSB_brm-1 (SEQ ID NO:3), BSB_mi-2-1 (SEQ ID NO:16), BSB_iswi-1-1 (SEQ ID NO:17), BSB_iswi-2-1 (SEQ ID NO:18), and BSB_chd1-1 (SEQ ID NO:19) were significantly different from that observed with the same amount of injected YFPv2 dsRNA (SEQ ID NO:5), with $p < 0.05$ (Table 4 and FIG. 4B). No eggs were produced by BSB_brm-1 and very few or none by BSB_mi-2-1 injected females. BSB_brm-1 (SEQ ID NO:3), BSB_mi-2-1 (SEQ ID NO:16), BSB_iswi-1-1 (SEQ ID NO:17), and BSB_chd1-1 (SEQ ID NO:19) dsRNA caused significant knockdown of transcript levels in the BSB ovary. FIG. 5. The transcript of BSB_iswi-2-1 (SEQ ID NO:18) was not readily detected by probe hydrolysis PCR.

The numbers of eggs hatched in the experiment below shows that the number of offspring produced from females injected with dsRNAs for BSB brahma, mi-2, iswi-1, iswi-2, and chd1 were significantly lower than the control. Table 5 and FIG. 4C. Egg hatch rates of BSB females injected with dsRNAs BSB_brm-1 (SEQ ID NO:3), BSB_mi-2-1 (SEQ ID NO:16), BSB_iswi-1-1 (SEQ ID NO:17), BSB_iswi-2-1 (SEQ ID NO:18), and BSB_chd1-1 (SEQ ID NO:19) were significantly different from those observed with the same amount of injected negative control YFPv2 dsRNA (SEQ ID NO:5), with $p < 0.05$ (Student's t-test).

TABLE 3

Brahma pRNAi: number of eggs oviposited per female per day. Ten females were injected with each dsRNA targeted against BSB brahma and negative control, YFPv2. Counts of oviposited eggs were collected starting on day 7 post injection, for up to 15 consecutive days. The N number of days during which eggs were collected varies between treatments due to female mortality impact of some dsRNAs. Means comparisons were performed with YFPv2 dsRNA as control, using a Student's t-test with Dunnett's adjustment in JMP® Pro 11.						
dsRNA	total # of eggs oviposited in 15 days	mean # of eggs/ day/ female	Std. Devia- tion	Std. Error	N days	T-test (p)
YFPv2	1280	8.66	1.84	0.48	15	
not inj.	1429	7.32	2.66	0.69	15	0.6697
BSB_brm-1	0	0	0	0	13	<0.0001*

*p-values < 0.05.

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TABLE 4

Oviposition by *E. heros* females injected with chromatin remodelers dsRNA. Total numbers of eggs oviposited in 15 days and average numbers of eggs per female injected with negative control YFPv2 dsRNA or chromatin remodeling ATPase dsRNAs. Twenty females were injected with each dsRNA. Egg counts started on day 9 post-injection and continued for 15 consecutive days. The N number of days during which eggs were collected varied between treatments due to female mortality in brm and mi-2 treatments. Means comparisons were performed on average numbers of eggs oviposited by females, using daily oviposition values. YFPv2 dsRNA was used as control for Student t-test with Dunnett's adjustment in JMP.

dsRNA	Total # of eggs in 15 days	Average # of eggs/ day/female	SEM	N Days	p-Value
YFPv2	1629	6.75	0.357	15	
BSB_brm-1	0	0.00	0.000	10	<0.0001*
BSB_chd1-1	496	2.65	0.338	15	<0.0001*
BSB_iswi-1-1	209	0.84	0.142	15	<0.0001*
BSB_iswi-2-1	1097	5.54	0.433	15	0.0171*
BSB_mi-2-1	42	0.22	0.085	13	<0.0001*

*significantly different from YFPv2 dsRNA $p < 0.05$.

N = number of days; SEM = standard error of the mean.

TABLE 5

Total and average numbers of eggs hatched from *E. heros* females injected with chromatin remodelers dsRNA. Total numbers of eggs hatched in 15-day collection and average number of eggs hatched per female per day of oviposition, from females injected with negative control YFPv2 dsRNA or chromatin remodeling dsRNAs. Twenty females were injected with each dsRNA. Nymph emergence was evaluated from eggs oviposited on day 9 post-injection for 15 consecutive days. Means comparisons were performed on numbers of eggs hatched each day per female, using daily values. YFPv2 dsRNA was used as the control for Student-t test with Dunnett's adjustment in JMP.

dsRNA	Total # of eggs hatched from 15-day collection	Average # of eggs hatched/ female/day	SEM	p-Value
YFPv2	1321	5.47	0.257	
BSB_brm-1	0	0.00	0.000	<0.0001*
BSB_chd1-1	51	0.28	0.054	<0.0001*
BSB_iswi-1-1	93	0.39	0.062	<0.0001*
BSB_iswi-2-1	312	1.63	0.253	<0.0001*
BSB_mi-2-1	34	0.17	0.067	<0.0001*

*significantly different from YFPv2 dsRNA $p < 0.05$.

SEM = standard error of the mean.

To determine the onset of pRNAi response, ovipositing females, 14 to 16 days post adult molt, were injected with BSB_brm-1 (SEQ ID NO:3) dsRNA. FIG. 6 shows that egg hatch was inhibited by day 4 post-injection (FIG. 6B) and oviposition halted by day 7 (FIG. 6A).

Based on the complete lack of oviposition in *E. heros* in response to brahma dsRNA and severe inhibition of oviposition in response to mi-2 dsRNA, we investigated the state of oocyte and ovary development in parent females. The females were examined 9 and 14 days post injection. By day nine after injection, control females began oviposition. Since brm dsRNA injections led to lethality within about two weeks, day 14 was chosen to capture phenotypes from the last surviving females. FIG. 4A. *E. heros* ovaries were dissected in 1×PBS under stereo microscope, and then fixed in 4% paraformaldehyde/1×PBS solution for 2 hours on ice. Trachea surrounding the ovaries was removed with #5 biology forceps. Images of three to four sets of ovaries for each treatment were captured with a Leica M205 FA stereo microscope (WETZLAR, Germany). Mature eggs and developing oocytes were observed in YFP dsRNA-injected

females. FIGS. 7C and D. Brahma and mi-2 dsRNA-injected females showed lack of ovary development and ovariole elongation. FIG. 7. These insects showed no maturing oocytes or mature eggs (FIGS. 7E, G, and H), or oocytes that were in a state of decay (FIG. 7F).

Contact with dsRNA molecules encoding sequences targeting SNF2-Helicase regions (SEQ ID NOs:34-37) and chromatin remodeling domains (SEQ ID NOs:38-41) by adult BSB females is demonstrates to a have surprising, dramatic and reproducible effect on egg viability. The mated females exposed to dsRNA produce a lower number of eggs than females exposed to untreated diet or diet treated with YFPv2 dsRNA.

The above results clearly document the systemic nature of RNAi in BSB adults, and the potential to achieve a parental effect where genes associated with embryonic development are knocked down in the eggs of females that are exposed to dsRNA. These observations confirm that the dsRNA can be taken up translocated to tissues (e.g., developing ovarioles) other than the point of contact (e.g., midgut or hemocoel).

The ability to knock down the expression of genes involved with embryonic development such that the eggs do not hatch, offers a unique opportunity to achieve and improve control of BSB. Because adults readily feed on above-ground reproductive tissues, adult BSB can be exposed to iRNA control agents by transgenic expression of dsRNA to achieve plant protection in the subsequent generation by preventing eggs from hatching. Delivery of the dsRNA through transgenic expression of dsRNA in plants, or by contact with surface-applied iRNAs, provides an important stacking partner for other transgenic approaches that target nymphs directly and enhance the overall durability of pest management strategies.

Example 6: Quantitive Real-Time PCR Analysis

E. heros tissues for qRT-PCR were collected from zero to three day-old females injected with dsRNA. After seven days, female ovaries were dissected under a stereo microscope in nuclease-free 1xPBS (pH 7.4) and frozen individually on dry ice in collection microtubes. Tissue disruption was performed with the RL lysis buffer and the Klecko™ tissue pulverizer (GARCIA MANUFACTURING, Visalia, Calif.). Following tissue maceration, the total RNA was

isolated in high throughput format using the Norgen® Total RNA Purification 96-well kit (NoRGEN BIOTEK CORP., Ontario, Canada) following the manufacturer's protocol using Turbo™ DNase (LIFE TECHNOLOGIES, Carlsbad, Calif.) for 1 hour at 37° C. on the elutant. cDNA synthesis was performed using the high capacity cDNA RT kit (LIFE TECHNOLOGIES, Carlsbad, Calif.) according to the manufacturer's protocol with the following modifications. Total RNA was adjusted to 50 ng/μL with nuclease-free water. RNA samples were heated to 70° C. for 10 minutes and cooled to 4° C. Half reactions were initiated by addition of 5 μL 2x mix. The primer mix, which is supplied solely as random primers, was first spiked with custom synthesized T₂₀VN oligo (INTEGRATED DNA TECHNOLOGIES, Coralville, Iowa) to a final concentration of 2 in order to improve the sensitivity of 3'UTR based assays. Following first strand synthesis, the samples were diluted 1:3 with nuclease-free water.

E. heros qRT-PCR primers and hydrolysis probes were designed using LightCycler® Probe Design Software 2.0 (ROCHE, Basel, Switzerland) for the reference gene and Primer Express® Software Version 3.0 (APPLIED BIOSYSTEMS, Grand Island, N.Y.) for the target genes. Table 6. Non-injected insects were used as controls. *E. heros* muscle actin (SEQ ID NO:73) was used as the reference gene. Probes were labeled with FAM (6-Carboxy Fluorescein Amidite). The final primer concentration was 0.4 μM, and the final probe concentration was 0.2 μM (in 10 reaction volumes). Relative transcript levels were analyzed by probe hydrolysis qRT-PCR using LightCycler®480. All assays included negative controls of no-template (mix only). For the standard curves, a blank was included in the source plate to check for sample cross-contamination. PCR cycling conditions included a 10 minute target activation incubation at 95° C., followed by 40 cycles of denaturation at 95° C. for 10 seconds, anneal/extend at 60° C. for 40 seconds, and FAM acquisition at 72° C. for 1 second. The reaction was followed by a 10 second cool-down at 40° C. *E. heros* iswi-2 was not detected reliably both in the negative controls and dsRNA exposed females, therefore iswi-2 data was omitted from the final results. The data was analyzed using LightCycler® Software v1.5 and relative changes in expression were calculated using 2^{-ΔΔC_T} method (Livak and Schmittgen (2001) Methods 25:402-8).

TABLE 6

Oligonucleotides and probes for BSB probe hydrolysis qPCR assay and primer efficacy results. MGB = Minor Groove Binder probes from Applied Biosystems.

Reference GENE	NAME	SEQUENCE	Product Length (bp)	Slope	Primer Efficiency (%)
Actin, muscle	Act42A-F	TCAAGGAAAACTGTGCTATGT (SEQ ID NO: 74)	120	-3.77	92
Actin, muscle	Act42A-R	TACCGATGGTGATGACCTGA (SEQ ID NO: 75)			
Actin, muscle	Act42A-FAM	ACCGCCGCTGCC (SEQ ID NO: 76)			
Target GENE	NAME	SEQUENCE			
brahma	brm-F	TCATCAAGGACAAGGCAGT (SEQ ID NO: 77)	205	-3.54	93.5

TABLE 6-continued

Oligonucleotides and probes for BSB probe hydrolysis qPCR assay and primer efficacy results. MGB = Minor Groove Binder probes from Applied Biosystems.					
brahma	brm-R	GACGGGAGGAGAAAGTTTAGA (SEQ ID NO: 78)			
brahma	brm-FAM	CGACGAGGGACACAGGATG (SEQ ID NO: 79)			
mi-2	mi-2-F	GATGAGGGCTTGCTGTT (SEQ ID NO: 80)	149	-3.55	95.5
mi-2	mi-2-R	GAGGCGGGAAGTATTGAC (SEQ ID NO: 81)			
mi-2	mi-2-FAM	ATGAGGAAGGAAGCAGAAGTGC (SEQ ID NO: 82)			
iswi-1	iswi-1-F	GAGTTCAACGAAGAAGACAGT AA (SEQ ID NO: 83)	155	-3.67	94.5
iswi-1	iswi-R	CGATGAGCACGATCCATAG (SEQ ID NO: 84)			
iswi-1	iswi-1-FAM	TTAGCCACCGCAGATGTAGTCA (SEQ ID NO: 85)			
iswi-2	iswi-2-F_MGB	ACGTAAGGGAGATGGATCTAT TTCA (SEQ ID NO: 86)	65	-3.96	89
iswi-2	iswi-2-R_MGB	CAGGGCTGCTTTTATCACTCT GT (SEQ ID NO: 87)			
iswi-2	iswi-2-FAM_MGB	CTCCACCTGTCTCTG (SEQ ID NO: 88)			
chd1	chd1-F	CAACAGTGGCTGGTCCTTCA (SEQ ID NO: 89)	68	-3.71	93
chd1	chd1-R	ACCAACTTGTGACATTGACGA AA (SEQ ID NO: 90)			
chd1	chd1-FAM	TCTGGTTTCAGCTCTT (SEQ ID NO: 91)			

Example 7: Construction of Plant Transformation Vectors

Entry vectors harboring a target gene construct for dsRNA hairpin formation comprising segments of one of various chromatin remodeling genes (SEQ ID NO:1 or SEQ ID NO:63 (brahma); SEQ ID NO:8 or SEQ ID NO:64 (BSB_mi-2); SEQ ID NO:10 or SEQ ID NO:65 (BSB_iswi-1); SEQ ID NO:12 or SEQ ID NO:66 (BSB_iswi-2); SEQ ID NO:14 or SEQ ID NO:67 (BSB_chd1); SEQ ID NO:30 (BSB_ino80); and SEQ ID NO:32 (BSB_domino)) are assembled using a combination of chemically synthesized fragments (DNA2.0, Menlo Park, Calif.) and standard molecular cloning methods. Intramolecular hairpin formation by RNA primary transcripts is facilitated by arranging (within a single transcription unit) two copies of a target gene segment in opposite orientation to one another, the two segments being separated by a linker sequence (e.g. ST-LS1 intron; Vancanneyt et al. (1990) Mol. Gen. Genet. 220:245-250). Thus, the primary mRNA transcript contains the two brahma or ortholog gene segment sequences as large inverted repeats of one another, separated by the linker sequence. A copy of a promoter (e.g. maize ubiquitin 1, U.S. Pat. No. 5,510,474; 35S from Cauliflower Mosaic Virus (CaMV); promoters from rice actin genes; ubiquitin promoters; pEMU; MAS; maize H3 histone promoter; ALS

promoter; phaseolin gene promoter; cab; rubisco; LAT52; Zm13; and/or apg) is used to drive production of the primary mRNA hairpin transcript, and a fragment comprising a 3' untranslated region, for example and without limitation, a maize peroxidase 5 gene (ZmPer5 3'UTR v2; U.S. Pat. No. 6,699,984), AtUbi10, AtEfl1, or StPinII is used to terminate transcription of the hairpin-RNA-expressing gene.

The entry vector described above is used in standard GATEWAY® recombination reactions with a typical binary destination vector to produce hairpin RNA expression transformation vectors for *Agrobacterium*-mediated plant embryo transformations.

A negative control binary vector which comprises a gene that expresses a YFP hairpin dsRNA is constructed by means of standard GATEWAY® recombination reactions with a typical binary destination vector and entry vector. The entry vector comprises a YFP hairpin sequence under the expression control of a maize ubiquitin 1 promoter and a fragment comprising a 3' untranslated region from a maize peroxidase 5 gene.

A binary destination vector comprises a herbicide tolerance gene (aryloxyalkanoate dioxygenase; (AAD-1 v3, U.S. Pat. No. 7,838,733, and Wright et al. (2010) Proc. Natl. Acad. Sci. U.S.A. 107:20240-5)) under the regulation of a plant operable promoter (e.g., sugarcane bacilliform badnavirus (ScBV) promoter (Schenk et al. (1999) Plant Mol.

Biol. 39:1221-30) or ZmUbi1 (U.S. Pat. No. 5,510,474)). 5' UTR and intron from these promoters, are positioned between the 3' end of the promoter segment and the start codon of the AAD-1 coding region. A fragment comprising a 3' untranslated region from a maize lipase gene (ZmLip 3'UTR; U.S. Pat. No. 7,179,902) is used to terminate transcription of the AAD-1 mRNA.

A further negative control binary vector that comprises a gene that expresses a YFP protein, is constructed by means of standard GATEWAY® recombination reactions with a typical binary destination vector and entry vector. The binary destination vector comprises a herbicide tolerance gene (aryloxyalkanoate dioxygenase; AAD-1 v3) (as above) under the expression regulation of a maize ubiquitin 1 promoter and a fragment comprising a 3' untranslated region from a maize lipase gene (ZmLip 3'UTR). The entry vector comprises a YFP coding region under the expression control of a maize ubiquitin 1 promoter and a fragment comprising a 3' untranslated region from a maize peroxidase 5 gene.

Example 8: Transgenic *Zea mays* Comprising Hemipteran Pest Sequences

Ten to 20 transgenic T₀ *Zea mays* plants harboring expression vectors for nucleic acids comprising a segment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67 are generated as described in EXAMPLE 5. A further 10-20 T₁ *Zea mays* independent lines expressing hairpin dsRNA for an RNAi construct are obtained for BSB challenge. Hairpin dsRNA may be derived comprising a segment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67. These are confirmed through RT-PCR or other molecular analysis methods. Total RNA preparations from selected independent T₁ lines are optionally used for RT-PCR with primers designed to bind in the linker of the hairpin expression cassette in each of the RNAi constructs. In addition, specific primers for each target gene in an RNAi construct are optionally used to amplify and confirm the production of the pre-processed mRNA required for siRNA production in planta. The amplification of the desired bands for each target gene confirms the expression of the hairpin RNA in each transgenic *Zea mays* plant. Processing of the dsRNA hairpin of the target genes into siRNA is subsequently optionally confirmed in independent transgenic lines using RNA blot hybridizations.

Moreover, RNAi molecules having mismatch sequences with more than 80% sequence identity to target genes affect hemipterans in a way similar to that seen with RNAi molecules having 100% sequence identity to the target genes. The pairing of mismatch sequence with native sequences to form a hairpin dsRNA in the same RNAi construct delivers plant-processed siRNAs capable of affecting the growth, development, reproduction, and viability of feeding hemipteran pests.

In planta delivery of dsRNA, siRNA, shRNA, hpRNA, or miRNA corresponding to target genes and the subsequent uptake by hemipteran pests through feeding results in down-regulation of the target genes in the hemipteran pest through RNA-mediated gene silencing. When the function of a target gene is important at one or more stages of development, the growth, development, and/or reproduction of the hemipteran pest is affected, and in the case of at least one of *Euschistus heros*, *Piezodorus guildinii*, *Halyomorpha halys*, *Nezara*

viridula, *Chinavia hilare*, *Euschistus serous*, *Dichelops melacanthus*, *Dichelops furcatus*, *Edessa meditabunda*, *Thyanta perditor*, *Chinavia marginatum*, *Horcias nobilellus*, *Taedia stigmosa*, *Dysdercus peruvianus*, *Neomegalotomus parvus*, *Leptoglossus zonatus*, *Niesthrea sidae*, or *Lygus lineolaris* leads to failure to successfully infest, feed, develop, and/or reproduce, or leads to death of the hemipteran pest. The choice of target genes and the successful application of RNAi is then used to control hemipteran pests.

Phenotypic Comparison of Transgenic RNAi Lines and Non-Transformed *Zea mays*.

Target hemipteran pest genes or sequences selected for creating hairpin dsRNA have no similarity to any known plant gene sequence. Hence it is not expected that the production or the activation of (systemic) RNAi by constructs targeting these hemipteran pest genes or sequences will have any deleterious effect on transgenic plants. However, development and morphological characteristics of transgenic lines are compared with non-transformed plants, as well as those of transgenic lines transformed with an "empty" vector having no hairpin-expressing gene. Plant root, shoot, foliage and reproduction characteristics are compared. There is no observable difference in root length and growth patterns of transgenic and non-transformed plants. Plant shoot characteristics such as height, leaf numbers and sizes, time of flowering, floral size and appearance are similar. In general, there are no observable morphological differences between transgenic lines and those without expression of target iRNA molecules when cultured in vitro and in soil in the glasshouse.

Example 9: Transgenic *Glycine max* Comprising Hemipteran Pest Sequences

Ten to 20 transgenic T₀ *Glycine max* plants harboring expression vectors for nucleic acids comprising a segment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67 are generated as is known in the art, including for example by *Agrobacterium*-mediated transformation, as follows. Mature soybean (*Glycine max*) seeds are sterilized overnight with chlorine gas for sixteen hours. Following sterilization with chlorine gas, the seeds are placed in an open container in a LAMINAR™ flow hood to dispel the chlorine gas. Next, the sterilized seeds are imbibed with sterile H₂O for sixteen hours in the dark using a black box at 24° C.

Preparation of Split-Seed Soybeans.

The split soybean seed comprising a portion of an embryonic axis protocol requires preparation of soybean seed material which is cut longitudinally, using a #10 blade affixed to a scalpel, along the hilum of the seed to separate and remove the seed coat, and to split the seed into two cotyledon sections. Careful attention is made to partially remove the embryonic axis, wherein about 1/2-1/3 of the embryo axis remains attached to the nodal end of the cotyledon.

Inoculation.

The split soybean seeds comprising a partial portion of the embryonic axis are then immersed for about 30 minutes in a solution of *Agrobacterium tumefaciens* (e.g., strain EHA 101 or EHA 105) containing binary plasmid comprising a segment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65,

SEQ ID NO:66, or SEQ ID NO:67. The *Agrobacterium tumefaciens* solution is diluted to a final concentration of $\lambda=0.6$ OD₆₅₀ before immersing the cotyledons comprising the embryo axis.

Co-Cultivation.

Following inoculation, the split soybean seed is allowed to co-cultivate with the *Agrobacterium tumefaciens* strain for 5 days on co-cultivation medium (*Agrobacterium Protocols*, vol. 2, 2nd Ed., Wang, K. (Ed.) Humana Press, New Jersey, 2006) in a Petri dish covered with a piece of filter paper.

Shoot Induction.

After 5 days of co-cultivation, the split soybean seeds are washed in liquid Shoot Induction (SI) media consisting of B5 salts, B5 vitamins, 28 mg/L Ferrous, 38 mg/L Na₂EDTA, 30 g/L sucrose, 0.6 g/L MES, 1.11 mg/L BAP, 100 mg/L TIMENTIN™, 200 mg/L cefotaxime, and 50 mg/L vancomycin (pH 5.7). The split soybean seeds are then cultured on Shoot Induction I (SII) medium consisting of B5 salts, B5 vitamins, 7 g/L Noble agar, 28 mg/L Ferrous, 38 mg/L Na₂EDTA, 30 g/L sucrose, 0.6 g/L MES, 1.11 mg/L BAP, 50 mg/L TIMENTIN™, 200 mg/L cefotaxime, 50 mg/L vancomycin (pH 5.7), with the flat side of the cotyledon facing up and the nodal end of the cotyledon imbedded into the medium. After 2 weeks of culture, the explants from the transformed split soybean seed are transferred to the Shoot Induction II (SI II) medium containing SI I medium supplemented with 6 mg/L glufosinate (LIBERTY®).

Shoot Elongation.

After 2 weeks of culture on SI II medium, the cotyledons are removed from the explants and a flush shoot pad containing the embryonic axis are excised by making a cut at the base of the cotyledon. The isolated shoot pad from the cotyledon is transferred to Shoot Elongation (SE) medium. The SE medium consists of MS salts, 28 mg/L Ferrous, 38 mg/L Na₂EDTA, 30 g/L sucrose and 0.6 g/L IVIES, 50 mg/L asparagine, 100 mg/L L-pyrogutamic acid, 0.1 mg/L IAA, 0.5 mg/L GA3, 1 mg/L zeatin riboside, 50 mg/L TIMENTIN™, 200 mg/L cefotaxime, 50 mg/L vancomycin, 6 mg/L glufosinate, 7 g/L Noble agar, (pH 5.7). The cultures are transferred to fresh SE medium every 2 weeks. The cultures are grown in a CONVIRON™ growth chamber at 24° C. with an 18 h photoperiod at a light intensity of 80-90 $\mu\text{mol}/\text{m}^2 \text{ sec}$.

Rooting.

Elongated shoots which developed from the cotyledon shoot pad are isolated by cutting the elongated shoot at the base of the cotyledon shoot pad, and dipping the elongated shoot in 1 mg/L IBA (Indole 3-butyric acid) for 1-3 minutes to promote rooting. Next, the elongated shoots are transferred to rooting medium (MS salts, B5 vitamins, 28 mg/L Ferrous, 38 mg/L Na₂EDTA, 20 g/L sucrose and 0.59 g/L IVIES, 50 mg/L asparagine, 100 mg/L L-pyrogutamic acid 7 g/L Noble agar, pH 5.6) in phyta trays.

Cultivation.

Following culture in a CONVIRON™ growth chamber at 24° C., 18 h photoperiod, for 1-2 weeks, the shoots which have developed roots are transferred to a soil mix in a covered sundae cup and placed in a CONVIRON™ growth chamber (models CMP4030 and CMP3244, Controlled Environments Limited, Winnipeg, Manitoba, Canada) under long day conditions (16 hours light/8 hours dark) at a light intensity of 120-150 $\mu\text{mol}/\text{m}^2 \text{ sec}$ under constant temperature (22° C.) and humidity (40-50%) for acclimatization of plantlets. The rooted plantlets are acclimated in sundae cups

for several weeks before they are transferred to the greenhouse for further acclimatization and establishment of robust transgenic soybean plants.

A further 10-20 T₁ *Glycine max* independent lines expressing hairpin dsRNA for an RNAi construct are obtained for BSB challenge. Hairpin dsRNA may be derived comprising a segment of SEQ ID NO:1, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:30, SEQ ID NO:32, SEQ ID NO:63, SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, or SEQ ID NO:67. These are confirmed through RT-PCR or other molecular analysis methods, as known in the art. Total RNA preparations from selected independent T₁ lines are optionally used for RT-PCR with primers designed to bind in the linker of the hairpin expression cassette in each of the RNAi constructs. In addition, specific primers for each target gene in an RNAi construct are optionally used to amplify and confirm the production of the pre-processed mRNA required for siRNA production in planta. The amplification of the desired bands for each target gene confirms the expression of the hairpin RNA in each transgenic *Glycine max* plant. Processing of the dsRNA hairpin of the target genes into siRNA is subsequently optionally confirmed in independent transgenic lines using RNA blot hybridizations.

RNAi molecules having mismatch sequences with more than 80% sequence identity to target genes affect BSB in a way similar to that seen with RNAi molecules having 100% sequence identity to the target genes. The pairing of mismatch sequence with native sequences to form a hairpin dsRNA in the same RNAi construct delivers plant-processed siRNAs capable of affecting the growth, development, reproduction, and viability of feeding hemipteran pests.

In planta delivery of dsRNA, siRNA, or miRNA corresponding to target genes and the subsequent uptake by hemipteran pests through feeding results in down-regulation of the target genes in the hemipteran pest through RNA-mediated gene silencing. When the function of a target gene is important at one or more stages of development, the growth, development, and/or reproduction of the hemipteran pest is affected, and in the case of at least one of *Euschistus heros*, *Piezodorus guildinii*, *Halyomorpha halys*, *Nezara viridula*, *Chinavia hilare*, *Euschistus servus*, *Dichelops melacanthus*, *Dichelops furcatus*, *Edessa meditabunda*, *Thyanta perditor*, *Chinavia marginatus*, *Horcias nobilellus*, *Taedia stigmata*, *Dysdercus peruvianus*, *Neomegalotomus parvus*, *Leptoglossus zonatus*, *Niesthrea sidae*, or *Lygus lineolaris* leads to failure to successfully infest, feed, develop, and/or reproduce, or leads to death of the hemipteran pest. The choice of target genes and the successful application of RNAi is then used to control hemipteran pests.

Phenotypic comparison of transgenic RNAi lines and non-transformed *Glycine max*. Target hemipteran pest genes or sequences selected for creating hairpin dsRNA have no similarity to any known plant gene sequence. Hence it is not expected that the production or the activation of (systemic) RNAi by constructs targeting these hemipteran pest genes or sequences will have any deleterious effect on transgenic plants. However, development and morphological characteristics of transgenic lines are compared with non-transformed plants, as well as those of transgenic lines transformed with an "empty" vector having no hairpin-expressing gene. Plant root, shoot, foliage and reproduction characteristics are compared. There is no observable difference in root length and growth patterns of transgenic and non-transformed plants. Plant shoot characteristics such as height, leaf numbers and sizes, time of flowering, floral size

and appearance are similar. In general, there are no observable morphological differences between transgenic lines and those without expression of target iRNA molecules when cultured in vitro and in soil in the glasshouse.

Example 10: *E. heros* Bioassays on Artificial Diet

In dsRNA feeding assays on artificial diet, 32-well trays are set up with an ~18 mg pellet of artificial diet and water, as for injection experiments. dsRNA at a concentration of 200 ng/ μ L is added to the food pellet and water sample, 100 μ L to each of two wells. Five 2nd instar *E. heros* nymphs are introduced into each well. Water samples and dsRNA that targets YFP transcript are used as negative controls. The experiments are repeated on three different days. Surviving insects are weighed and the mortality rates are determined after 7 days of treatment.

Feeding bioassays on adult female *E. heros* are performed as 32-well trays as described above. Young (less than one week of adulthood) mated females are introduced into bioassay trays with artificial diet, one per tray. After 7 days of exposure to dsRNA up to ten adult females are moved to containers with green beans, water, seeds, and two males. Female viability as well as the numbers of eggs oviposited and eggs hatched are recorded for the following two weeks. The data shows that the numbers of eggs oviposited and/or hatched are significantly reduced.

Example 11: Transgenic *Arabidopsis thaliana* Comprising Hemipteran Pest Sequences

Arabidopsis transformation vectors containing a target gene construct for hairpin formation comprising segments of BSB_brahma (SEQ ID NO:1 or SEQ ID NO:63), BSB_mi-2 (SEQ ID NO:8 or SEQ ID NO:64), BSB_iswi-1 (SEQ ID NO:10 or SEQ ID NO:65), BSB_iswi-2 (SEQ ID NO:12 or SEQ ID NO:66), BSB_chd1 (SEQ ID NO:14 or SEQ ID NO:67), BSB_ino80 (SEQ ID NO:30), and/or BSB_domino (SEQ ID NO:32) are generated using standard molecular methods similar to EXAMPLE 5. *Arabidopsis* transformation is performed using standard *Agrobacterium*-based procedure. T₁ seeds are selected with glufosinate tolerance selectable marker. Transgenic T₁ *Arabidopsis* plants are generated and homozygous simple-copy T2 transgenic plants are generated for insect studies. Bioassays are performed on growing *Arabidopsis* plants with inflorescences. Five to ten insects are placed on each plant and monitored for survival within 14 days.

Construction of *Arabidopsis* Transformation Vectors.

Entry clones based on an entry vector harboring a target gene construct for hairpin formation comprising a segment of BSB_brahma (SEQ ID NO:1 or SEQ ID NO:63), BSB_mi-2 (SEQ ID NO:8 or SEQ ID NO:64), BSB_iswi-1 (SEQ ID NO:10 or SEQ ID NO:65), BSB_iswi-2 (SEQ ID NO:12 or SEQ ID NO:66), BSB_chd1 (SEQ ID NO:14 or SEQ ID NO:67), BSB_ino80 (SEQ ID NO:30), and/or BSB_domino (SEQ ID NO:32) are assembled using a combination of chemically synthesized fragments (DNA2.0, Menlo Park, Calif.) and standard molecular cloning methods. Intramolecular hairpin formation by RNA primary transcripts is facilitated by arranging (within a single transcription unit) two copies of a target gene segment in opposite orientations, the two segments being separated by an linker sequence (e.g. ST-LS1 intron) (Vancanney et al. (1990) Mol. Gen. Genet. 220(2):245-50). Thus, the primary mRNA transcript contains the two chromatin remodeling gene segment sequences as large inverted repeats of one

another, separated by the linker sequence. A copy of a promoter (e.g. *Arabidopsis thaliana* ubiquitin 10 promoter (Callis et al. (1990) J. Biological Chem. 265:12486-12493)) is used to drive production of the primary mRNA hairpin transcript, and a fragment comprising a 3' untranslated region from Open Reading Frame 23 of *Agrobacterium tumefaciens* (AtuORF23 3' UTR v1; U.S. Pat. No. 5,428,147) is used to terminate transcription of the hairpin-RNA-expressing gene.

The hairpin clone within the entry vector described above is used in standard GATEWAY® recombination reaction with a typical binary destination vector to produce hairpin RNA expression transformation vectors for *Agrobacterium*-mediated *Arabidopsis* transformation.

The binary destination vector comprises a herbicide tolerance gene, DSM-2v2 (U.S. Patent App. No. 2011/0107455), under the regulation of a Cassava vein mosaic virus promoter (CsVMV Promoter v2, U.S. Pat. No. 7,601,885; Verdager et al. (1996) Plant Mol. Biol. 31:1129-39). A fragment comprising a 3' untranslated region from Open Reading Frame 1 of *Agrobacterium tumefaciens* (AtuORF1 3' UTR v6; Huang et al. (1990) J. Bacteriol. 172:1814-22) is used to terminate transcription of the DSM2v2 mRNA.

A negative control binary construct, which comprises a gene that expresses a YFP hairpin RNA, is constructed by means of standard GATEWAY® recombination reactions with a typical binary destination vector and entry vector. An entry construct comprises a YFP hairpin sequence (hpYFP v2, SEQ ID NO:42) under the expression control of an *Arabidopsis* Ubiquitin 10 promoter (as above) and a fragment comprising an ORF23 3' untranslated region from *Agrobacterium tumefaciens* (as above).

Production of Transgenic *Arabidopsis* Comprising Insecticidal Hairpin RNAs: *Agrobacterium*-Mediated Transformation.

Binary plasmids containing hairpin sequences are electroporated into an *Agrobacterium* strain. The recombinant *Agrobacterium* clones are confirmed by restriction analysis of plasmids preparations of the recombinant *Agrobacterium* colonies. A Qiagen Plasmid Max Kit (Qiagen, Cat#12162) is used to extract plasmids from *Agrobacterium* cultures following the manufacture recommended protocol.

Arabidopsis Transformation and T₁ Selection.

Twelve to fifteen *Arabidopsis* plants (c.v. Columbia) are grown in 4" pots in the green house with light intensity of 250 μ mol/m², 25° C., and 18:6 hours of light:dark conditions. Primary flower stems are trimmed one week before transformation. *Agrobacterium* inoculums are prepared by incubating 10 μ L recombinant *Agrobacterium* glycerol stock in 100 mL LB broth (Sigma L3022)+100 mg/L Spectinomycin+50 mg/L Kanamycin at 28° C. and shaking at 225 rpm for 72 hours. *Agrobacterium* cells are harvested and suspended into 5% sucrose+0.04% Silwet-L77 (Lehle Seeds Cat # VIS-02)+10 μ g/L benzamino purine (BA) solution to OD₆₀₀ 0.8–1.0 before floral dipping. The above-ground parts of the plant are dipped into the *Agrobacterium* solution for 5-10 minutes, with gentle agitation. The plants are then transferred to the greenhouse for normal growth with regular watering and fertilizing until seed set.

Example 12: Growth and Bioassays of Transgenic *Arabidopsis*

Selection of T₁ *Arabidopsis* Transformed with Hairpin RNAi Constructs.

Up to 200 mg of T₁ seeds from each transformation are stratified in 0.1% agarose solution. The seeds are planted in

germination trays (10.5"x21"x1"; T.O. Plastics Inc., Clearwater, Minn.) with #5 sunshine media. Transformants are selected for tolerance to Ignite® (glufosinate) at 280 g/ha at 6 and 9 days post planting. Selected events are transplanted into 4" diameter pots. Insertion copy analysis is performed within a week of transplanting via hydrolysis quantitative Real-Time PCR (qPCR) using Roche LightCycler480™. The PCR primers and hydrolysis probes are designed against DSM2v2 selectable marker using LightCycler™ Probe Design Software 2.0 (Roche). Plants are maintained at 24° C., with a 16:8 hour light:dark photoperiod under fluorescent and incandescent lights at intensity of 100-150 mE/m² s.

E. heros Nymph Plant Feeding Bioassay.

At least four low copy (1-2 insertions), four medium copy (2-3 insertions), and four high copy (≥4 insertions) events are selected for each construct. Plants are grown to a reproductive stage (plants containing flowers and siliques). The surface of soil is covered with ~50 mL volume of white sand for easy insect identification. Five to ten 2nd instar *E. heros* nymphs are introduced onto each plant. The plants are covered with plastic tubes that are 3" in diameter, 16" tall, and with wall thickness of 0.03" (Item No. 484485, Visipack Fenton Mo.); the tubes are covered with nylon mesh to isolate the insects. The plants are kept under normal temperature, light, and watering conditions in a conviron. In 14 days, the insects are collected and weighed; percent mortality as well as growth inhibition (1-weight treatment/weight control) are calculated. YFP hairpin-expressing plants are used as controls.

The pRNAi *Arabidopsis* T₁ plants are selected and grown in greenhouse, as described above. One to 5 newly emerged BSB adults are released on each plant and the entire plant is covered as described above to prevent adults from escaping. One week after release, female adults are recovered from each plant and maintained in the laboratory for egg collection. Depending on the parental RNAi target and expected phenotype, parameters such as number of eggs per female, percent egg hatch and nymph mortality are recorded and compared with control plants.

T₂ *Arabidopsis* Seed Generation and T₂ Bioassays.

T₂ seed is produced from selected low copy (1-2 insertions) events for each construct. Plants (homozygous and/or heterozygous) are subjected to *E. heros* nymph and adult feeding bioassay, as described above. T₃ seed is harvested from homozygotes and stored for future analysis.

Example 13: Transformation of Additional Crop Species

Cotton is transformed with brahma, mi-2, iswi-1, iswi-2, chd1, ino80, and/or domino (with or without a chloroplast transit peptide) to provide control of stink bugs by utilizing a method known to those of skill in the art, for example, substantially the same techniques previously described in EXAMPLE 14 of U.S. Pat. No. 7,838,733, or Example 12 of PCT International Patent Publication No. WO 2007/053482.

Example 14: pRNAi-Mediated Insect Protection

Parental RNAi that causes egg mortality or loss of egg viability brings further durability benefits to transgenic crops that use RNAi and other mechanisms for insect protection. A basic two-patch model was used to demonstrate this utility.

One patch contained a transgenic crop expressing insecticidal ingredients, and the second patch contained a refuge crop not expressing insecticidal ingredients. Eggs were

oviposited in the two modeled patches according to their relative proportions. In this example, the transgenic patch represented 95% of the landscape, and the refuge patch represented 5%. The transgenic crop expressed an insecticidal protein active against the insect.

Pest resistance to the insecticidal protein was modeled as monogenic, with two possible alleles; one (S) conferring susceptibility, and the other (R) conferring resistance. The insecticidal protein was modeled to cause 97% mortality of homozygous susceptible (SS) nymphs that feed on it. There was assumed to be no mortality of nymphs that are homozygous for the resistance allele (RR). Resistance to the insecticidal protein was assumed to be incompletely recessive, whereby the functional dominance is 0.3 (there is 67.9% mortality of nymphs that are heterozygous (RS) for resistance to the protein that feed on the transgenic crop).

The transgenic crop also expressed parentally active dsRNA that, through RNA-interference (pRNAi), causes the eggs of adult female insects that are exposed to the transgenic crop to be non-viable. Insect resistance to the pRNAi was also considered to be monogenic with two possible alleles; one (X) conferring susceptibility of the adult female to RNAi, and the other (Y) conferring resistance of the adult female to RNAi. Assuming a high level of exposure to the dsRNAs, the pRNAi was modeled to cause 99.9% of eggs produced by a homozygous susceptible (XX) female to be non-viable. The model assumed that pRNAi has no effect on the viability of eggs produced by homozygous resistant (YY) females. Resistance to the dsRNA was assumed to be recessive, whereby the functional dominance is 0.01 (98.9% of eggs produced by a female that is heterozygous (XY) for resistance to dsRNA are non-viable).

In the model, there was random mating among surviving adults and random oviposition across the two patches in accordance with their relative proportions. The genotypic frequencies of viable offspring followed Mendelian genetics for a two-locus genetic system.

The effect of pRNAi required the adult females to feed on plant tissue expressing parental active dsRNA. The interference with egg development may be lower for adult females emerging from the refuge crop than from the transgenic crop; adults are expected to feed more extensively in the patch in which they emerged following nymph development. Therefore, the relative magnitude of the pRNAi effect on female adults emerging from the refuge patch was varied, with the proportion of the pRNAi effect ranging from 0 (no effect of pRNAi on adult females emerging from the refuge patch) to 1 (same effect of pRNAi on adult females emerging from the refuge patch as on adult females emerging from the transgenic patch).

This model could be easily adjusted to demonstrate the situation when the effect of pRNAi is also or alternatively achieved by feeding of adult males on plant tissue expressing parental active dsRNA.

Frequencies of the two resistance alleles were calculated across generations. The initial frequencies of both of the resistance alleles (R and Y) were assumed to be 0.005. Results were presented as the number of insect generations for the frequencies of each of the resistance alleles to reach 0.05. To examine the resistance delay caused by the pRNAi, simulations that included pRNAi were compared to simulations that did not include pRNAi, but were identical in every other way. FIG. 8.

The model was also modified to include nymph-active interfering dsRNA in combination with the BSB-active insecticidal protein in the transgenic crop. Therein, the nymph RNAi was assigned an effect of 97% nymph mor-

ality for homozygous RNAi-susceptible nymphs (genotype XX), and no effect on nymphs that are homozygous RNAi-resistant (YY). There was 67.9% mortality of nymphs that were heterozygous for RNAi-resistance (XY). It was assumed that the same mechanism of resistance applied to both nymph active RNAi and pRNAi. As before, the pRNAi effect on adult females emerging from the refuge patch relative to the effect on adult females emerging from the transgenic patch was varied from 0 to 1. As before, to examine the resistance delay caused by the pRNAi, simulations that included pRNAi were compared to simulations that did not include pRNAi, but were identical in every other way (including nymph RNAi). FIG. 9.

A clear resistance management benefit of pRNAi was observed when the magnitude of the pRNAi effect on egg

viability for female adults emerging from the refuge patch was reduced compared with magnitude of the effect for adults emerging from the transgenic patch. The transgenic crops that produced parental active dsRNA in addition to an insecticidal protein were much more durable compared with transgenic crops that produced only an insecticidal protein. Similarly, transgenic crops that produced parental active dsRNA in addition to both an insecticidal protein and a nymph active dsRNA were much more durable compared with transgenic crops that produced only an insecticidal protein and a larval active dsRNA. In the latter case, the durability benefit applied to both the insecticidal protein and the insecticidal interfering dsRNA.

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<210> SEQ ID NO 2

<211> LENGTH: 1523

<212> TYPE: PRT

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 2

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Gln Gly Ala Pro Ser Pro Met Pro Pro Ser Asn Gln Gln Ala Ala Ser
35           40           45

Pro Met Gly Pro Pro His His Pro His Ser Pro Thr Gly Tyr Gln Gly
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Gly Met Pro His Met Asn Gly Pro Asn Gly Val Pro Pro Gly Met Gln
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Gln Ala Thr Gln Thr Phe Gln Pro His Gln Gln Leu Pro Pro His Gln
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Gln Pro Pro Met Gln Thr Ala Pro Gly Gly Pro Ala Ser Gly Gly Gly
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Gln Glu Asn Leu Ser Ala Leu Gln Arg Ala Ile Asp Ser Met Glu Glu
115          120          125

Lys Gly Leu Gln Glu Asp Pro Arg Tyr Ser Gln Leu Leu Ala Leu Arg
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Ala Arg His Ala Asn Met Glu Pro Pro Val Arg Pro Pro Ser Gln Leu
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Val Gly Gly Gly Phe Ser Gly Glu Gly Gly Ala Pro Pro Pro Ala Lys
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His Ser Phe Ser Ala Asn Gln Leu Gln Gln Leu Arg Val Gln Ile Met

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Phe	Lys	Glu	Phe	His	Arg	Asn	Asn	Val	Ala	Lys	Val	Gly	Arg	Leu	Asn		
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Ala	Lys	Leu	His	Trp	Lys	Tyr	Met	Ile	Ile	Asp	Glu	Gly	His	Arg	Met 830
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Glu	Val	Glu	Ser	Gln	Leu	Pro	Glu	Lys	Ile	Glu	Tyr	Ile	Val	Lys	Cys 940
Asp 945	Met	Ser	Gly	Leu	Gln	Arg	Val	Leu	Tyr	Arg	His	Met	Gln	Ser	Lys 960
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Ala Arg 1220	Thr Glu	Asp Glu	Phe 1225	Asn Leu	Phe Gln	Lys 1230	Ile Asp	Leu
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Arg Leu 1250	Val Glu	Glu Ala	Glu 1255	Leu Pro	Asp Trp	Leu 1260	Val Lys	Asn
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Ser Leu 1295	Thr Glu	Lys Glu	Trp 1300	Leu Lys	Ala Ile	Asp 1305	Asp Asn	Val
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Arg Gly 1325	Lys Arg	Arg Arg	Arg 1330	Gly Glu	Asp Asp	Glu 1335	Glu Asp	Ala
Ser Thr 1340	Ser Lys	Arg Arg	Lys 1345	Tyr Ser	Pro Ser	Glu 1350	Asn Lys	Leu
Arg Arg 1355	Arg Met	Arg Asn	Leu 1360	Met Asn	Ile Val	Val 1365	Lys Tyr	Thr
Asp Ser 1370	Asp Ser	Arg Val	Leu 1375	Ser Glu	Pro Phe	Met 1380	Lys Leu	Pro
Ser Arg 1385	His Lys	Tyr Pro	Asp 1390	Tyr Tyr	Glu Leu	Ile 1395	Lys Lys	Pro
Ile Asp 1400	Ile Lys	Arg Ile	Leu 1405	Ala Lys	Val Glu	Glu 1410	Cys Lys	Tyr
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Ser Ile Val Leu Glu Ser Val Phe Ser Asn Ala Arg Gln Lys Val		
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Glu Gln Asp Asn Asp Ser Asp Asp Asp Glu Ser Lys Gly Asp Gln		
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Glu Asp Ala Ala Ser Asp Thr Ser Ser Val Lys Met Lys Leu Lys		
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Leu Lys Pro Gly Arg Thr Arg Gly Ser Gly Ala Gly Gly Lys Arg		
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<210> SEQ ID NO 4
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<400> SEQUENCE: 4

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<210> SEQ ID NO 5
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<400> SEQUENCE: 5

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<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer YFPv2-F

<400> SEQUENCE: 6

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<211> LENGTH: 46

<212> TYPE: DNA

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<223> OTHER INFORMATION: Primer YFPv2-R

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<211> LENGTH: 6346

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<213> ORGANISM: *Euchistus heros*

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tctaattggcg tctgaagaag aagttgacga gtgtttacca gttgacgatg aagttgacac 180

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<210> SEQ ID NO 9
<211> LENGTH: 1938
<212> TYPE: PRT
<213> ORGANISM: Euchistus heros

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<400> SEQUENCE: 9

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Pro Glu Asp Ala Arg Lys Lys Lys Lys Gly Lys Lys Arg Lys Ala Lys
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Gly Glu Ser Lys Lys Glu Lys Lys Arg Lys Lys Arg Lys Lys Asn Asp
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Ser Ala Glu Glu Ser Glu Gly Gly Gly Glu Glu Glu Gly Asp Ser Asp
85         90         95
Tyr Gly Arg Lys Ser Lys Lys Ser Lys Gly Thr Ser Gln Pro Lys Pro
100        105        110
Val Gln Gln Asp Ser Ser Gly Gly Val Pro Ser Val Glu Glu Val Cys
115        120        125
Ser Leu Phe Gly Leu Thr Asp Val Gln Ile Asp Tyr Thr Glu Asp Asp
130        135        140
Tyr Gln Asn Leu Thr Thr Tyr Lys Leu Phe Gln Gln His Val Arg Pro
145        150        155        160
Ile Leu Ala Lys Asp Asn Gln Lys Val Pro Ile Gly Lys Met Met Met
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Leu Val Ala Ala Lys Trp Arg Asp Phe Cys Asn Ser Asn Pro Asn Ala
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Gln Gln Glu Pro Asp Pro Glu Ala Ser Glu Glu Gln Glu Tyr Ser Lys
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Pro Thr Arg Thr Arg Pro Ser Arg Val Ser Thr Thr Gln Asn Asp Asp
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Glu Glu Asp Asp Asp Ala Asp Glu Arg Gly Arg Lys Lys Arg Ser Gly
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Lys Val Pro Thr Leu Lys Ile Lys Ile Gly Lys Arg Lys Gln Asn Ser
260        265        270
Asp Glu Glu Asp Glu Gly Ser Val Gly Ala Val Ser Glu Arg Asp Ser
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Asp Ala Glu Phe Glu Gln Met Leu Ala Glu Ala Glu Glu Val Asn Lys
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Pro Glu Gly Val Val Glu Glu Glu Glu Gly Ala Glu Val Ala Pro Val
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Ile	Leu	Cys	Asp	Thr	Cys	Pro	Arg	Ala	Tyr	His	Leu	Val	Cys	Leu	Asp
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Cys	Arg	Val	Cys	Lys	Asp	Gly	Gly	Glu	Leu	Leu	Cys	Cys	Asp	Ser	Cys
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Gly	Lys	Val	Ser	Lys	Ile	Leu	Thr	Trp	Arg	Trp	Leu	Glu	Ser	Pro	Ser
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Trp	Val	Ser	Glu	Leu	Gln	Met	Asp	Val	Phe	His	Thr	Gln	Met	Ile	Arg
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Ser	Tyr	Ile	Arg	Lys	Tyr	Asp	Met	Asp	Glu	Pro	Pro	Lys	Leu	Glu	Glu
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Pro	Leu	Asp	Glu	Ala	Asp	Asn	Arg	Met	Lys	Arg	Ile	Arg	Glu	Ala	Asn
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Ile	Asn	Glu	Gln	Glu	Leu	Glu	Glu	Lys	Tyr	Tyr	Lys	Tyr	Gly	Ile	Lys
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Ala	Ile	Glu	Tyr	Tyr	Asn	Glu	Met	Arg	Ala	Cys	Cys	Leu	Gly	Glu	Ser
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Lys	Lys	Leu	Lys	Lys	Gly	Lys	Gly	Lys	Arg	Ser	Lys	Arg	Asp	Gln	Asp
				645					650					655	
Asp	Glu	Glu	Gly	Ser	Arg	Ser	Ala	Gly	Met	Met	Gly	Val	Gly	Gly	Pro
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Ala	Thr	Gly	Gln	Tyr	Phe	Pro	Pro	Pro	Glu	Lys	Pro	Val	Thr	Asp	Leu
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Lys	Lys	Lys	Tyr	Asp	Lys	Gln	Pro	Asp	Tyr	Leu	Asp	Val	Ser	Gly	Met
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Cys	Leu	His	Pro	Tyr	Gln	Leu	Glu	Gly	Leu	Asn	Trp	Leu	Arg	Tyr	Ser
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Trp	Gly	Gln	Gly	Thr	Asp	Thr	Ile	Leu	Ala	Asp	Glu	Met	Gly	Leu	Gly
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Thr Tyr Val Gly Asp Lys Asp Ser Arg Ala Val Ile Arg Glu Asn Glu 785 790 795 800		
Phe Ser Phe Asp Asp Asn Ala Val Arg Gly Gly Arg Gly Val Ser Lys 805 810 815		
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Leu Ile Ser Ile Asp Val Thr Cys Leu Gly Ser Ile Glu Trp Ala Val 835 840 845		
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Phe Ala Asp Ile Ser Lys Glu Glu Gln Val Lys Arg Leu His Glu Leu 915 920 925		
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1295			1300			1305		
Lys Gln	Leu Tyr	Lys Val	Asn	Tyr Ile	Asp Gly	Gly	Val Met	Asp
1310			1315			1320		
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1535			1540			1545		

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<210> SEQ ID NO 10
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 <213> ORGANISM: *Euchistus heros*

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gattcggatt ggaacctca aatggatctc caggctatgg atcgtgctca tcgtattggt   1800
caaaagaaac aagtcaaagt gttcaggatg ataactgaaa acacagtga agagaaaatt   1860
gttgagagag ctgaaataaa actccgcctc gataagttgg tcatccaaca aggcaggctg   1920
gtagacaata aaacggcact caacaaagat gaaatgttga atatgatccg tcacggtgcc   1980
aatcatgtat ttgccagtaa agattctgaa atcaccgatg aagacattga cactattttg   2040
gaaaaaggcg aagcaaggac ggaagaaatg aataaaaaac ttgaacaact cggtgattct   2100
aatttgaaag acttcatgat ggaaccccg actgagtcag tttaccaatt cgaaggagag   2160

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gattacaggg aaaagcagaa agtttttagga ataggaagtt ggatagaacc tccaaaaaga 2220
gaacgtaaag ctaattacgc tgcgatgcc tatttttaggg aagcattgag agtatcagaa 2280
cctaagctc ccaaggcacc gaggcctcct aaacagccta tagttcaaga ttccaattc 2340
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ggctacaaag ttctaaaaa tctgaatta ggttctgatg catcacgtgt ccaaagga 2460
gaacaaagaa agatagatga ggcagaacct ttatcagaag aagaactcgc tgaaaaggaa 2520
aaacttctta cgcagggttt taccaattgg actaaaagag atttcaacca gtttattaaa 2580
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gacatagatc gtatcatggg gcagatcgac aggggagagg ctaaaattca aaggagagca 2760
agtattaaga aagctctcga tacaagatg agccggtaca gagccccatt tcatcaactt 2820
cgcatctcct acggtacgaa taagggttaag aactataccg aggaagaaga tagattcctt 2880
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atggtcaggt gtgcgcctca gttcagattc gactgggtca tcaaatcgag aacagccatg 3000
gaattgcaga ggcgttgtaa tactctaatt actctcatcg aaagagaaaa tcaggaaactt 3060
gaggagaggg aaagagccga gaagaggaaa ggaagaggaa gtgggcgtgg tctgtgttc 3120
ggtaaaagga aaggagacgg ttccatttca tctccccctc ctgtccctgg ccaaggggat 3180
aagaacagcc ccgccagaaa aaagaaaaaa atgtagtttc acctctcat gaaaggaact 3240
cattttaaga tatctttttc tagatattta ttttgtgaaa actgtgatgt attttatct 3300
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tagccccctc acccccctaat aattcataaa t 3391

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<210> SEQ ID NO 11
<211> LENGTH: 1023
<212> TYPE: PRT
<213> ORGANISM: Euchistus heros

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<400> SEQUENCE: 11

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```

Met Ser Lys Pro Asn Glu Val Ser Leu Asp Thr Thr Asp Thr Val Glu
1      5      10      15
Ile Ser Asn Glu Ser Ser Gly Asp Thr Glu Ser Ser Lys Gly Lys Asn
20     25     30
Glu Asp Phe Glu Thr Lys Ile Glu Thr Asp Arg Ser Arg Arg Phe Glu
35     40     45
Phe Leu Leu Lys Gln Thr Glu Ile Phe Ser His Phe Met Thr Asn Gln
50     55     60
Gly Lys Ser Asn Ser Pro Ala Lys Pro Lys Val Gly Arg Pro Arg Lys
65     70     75     80
Glu Thr Asn Lys Leu Ala Pro Ala Gly Gly Asp Gly Ser Ala Asp His
85     90     95
Arg His Arg Met Thr Glu Gln Glu Glu Asp Glu Glu Leu Leu Ala Glu
100    105    110
Ser Asn Thr Ser Ser Lys Ser Leu Ala Arg Phe Asp Ala Ser Pro Phe
115    120    125
Tyr Ile Lys Ser Gly Glu Leu Arg Asp Tyr Gln Ile Arg Gly Leu Asn
130    135    140
Trp Met Ile Ser Leu Tyr Glu His Gly Ile Asn Gly Ile Leu Ala Asp
145    150    155    160

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Glu Met Gly Leu Gly Lys Thr Leu Gln Thr Ile Ser Leu Leu Gly Tyr	165	170	175
Met Lys His Tyr Arg Asn Ile Pro Gly Pro His Met Val Ile Val Pro	180	185	190
Lys Ser Thr Leu Ala Asn Trp Met Asn Glu Phe Lys Lys Trp Cys Pro	195	200	205
Thr Leu Arg Ala Val Cys Leu Ile Gly Asp Gln Glu Thr Arg Asn Ala	210	215	220
Phe Ile Arg Asp Thr Leu Met Pro Gly Glu Trp Asp Val Cys Val Thr	225	230	235
Ser Tyr Glu Met Ile Ile Arg Glu Lys Ser Val Phe Lys Lys Phe Asn	245	250	255
Trp Arg Tyr Met Val Ile Asp Glu Ala His Arg Ile Lys Asn Glu Lys	260	265	270
Ser Lys Leu Ser Glu Ile Val Arg Glu Phe Lys Thr Thr Asn Arg Leu	275	280	285
Leu Leu Thr Gly Thr Pro Leu Gln Asn Asn Leu His Glu Leu Trp Ser	290	295	300
Leu Leu Asn Phe Leu Leu Pro Asp Val Phe Asn Ser Ser Asp Asp Phe	305	310	315
Asp Ser Trp Phe Asn Thr Asn Thr Phe Leu Gly Asp Asn Ser Leu Val	325	330	335
Glu Arg Leu His Ala Val Leu Arg Pro Phe Leu Leu Arg Arg Leu Lys	340	345	350
Ser Glu Val Glu Lys Lys Leu Lys Pro Lys Lys Glu Val Lys Ile Tyr	355	360	365
Val Gly Leu Ser Lys Met Gln Arg Glu Trp Tyr Thr Lys Val Leu Met	370	375	380
Lys Asp Ile Asp Ile Val Asn Gly Ala Gly Arg Val Glu Lys Met Arg	385	390	395
Leu Gln Asn Ile Leu Met Gln Leu Arg Lys Cys Ser Asn His Pro Tyr	405	410	415
Leu Phe Asp Gly Ala Glu Pro Gly Pro Pro Tyr Ser Thr Asp Glu His	420	425	430
Leu Val Tyr Asn Ser Gly Lys Met Val Ile Leu Asp Lys Leu Leu Pro	435	440	445
Lys Leu Gln Glu Gln Gly Ser Arg Val Leu Val Phe Ser Gln Met Thr	450	455	460
Arg Met Ile Asp Ile Leu Glu Asp Tyr Cys Tyr Trp Arg Gly Tyr Asn	465	470	475
Tyr Cys Arg Leu Asp Gly Asn Thr Pro His Glu Asp Arg Gln Arg Gln	485	490	495
Ile Asn Glu Phe Asn Glu Glu Asp Ser Lys Lys Phe Ile Phe Met Leu	500	505	510
Ser Thr Arg Ala Gly Gly Leu Gly Ile Asn Leu Ala Thr Ala Asp Val	515	520	525
Val Ile Leu Tyr Asp Ser Asp Trp Asn Pro Gln Met Asp Leu Gln Ala	530	535	540
Met Asp Arg Ala His Arg Ile Gly Gln Lys Lys Gln Val Lys Val Phe	545	550	555
Arg Met Ile Thr Glu Asn Thr Val Glu Glu Lys Ile Val Glu Arg Ala	565	570	575

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Glu	Ile	Lys	Leu	Arg	Leu	Asp	Lys	Leu	Val	Ile	Gln	Gln	Gly	Arg	Leu
			580					585					590		
Val	Asp	Asn	Lys	Thr	Ala	Leu	Asn	Lys	Asp	Glu	Met	Leu	Asn	Met	Ile
		595					600					605			
Arg	His	Gly	Ala	Asn	His	Val	Phe	Ala	Ser	Lys	Asp	Ser	Glu	Ile	Thr
	610					615					620				
Asp	Glu	Asp	Ile	Asp	Thr	Ile	Leu	Glu	Lys	Gly	Glu	Ala	Arg	Thr	Glu
	625				630					635					640
Glu	Met	Asn	Lys	Lys	Leu	Glu	Gln	Leu	Gly	Asp	Ser	Asn	Leu	Lys	Asp
				645					650					655	
Phe	Met	Met	Glu	Thr	Pro	Thr	Glu	Ser	Val	Tyr	Gln	Phe	Glu	Gly	Glu
			660					665					670		
Asp	Tyr	Arg	Glu	Lys	Gln	Lys	Val	Leu	Gly	Ile	Gly	Ser	Trp	Ile	Glu
		675					680					685			
Pro	Pro	Lys	Arg	Glu	Arg	Lys	Ala	Asn	Tyr	Ala	Val	Asp	Ala	Tyr	Phe
	690					695					700				
Arg	Glu	Ala	Leu	Arg	Val	Ser	Glu	Pro	Lys	Ala	Pro	Lys	Ala	Pro	Arg
	705				710					715					720
Pro	Pro	Lys	Gln	Pro	Ile	Val	Gln	Asp	Phe	Gln	Phe	Phe	Pro	Pro	Arg
				725					730					735	
Leu	Phe	Glu	Leu	Leu	Asp	Gln	Glu	Ile	Tyr	Tyr	Phe	Arg	Lys	Thr	Val
			740					745					750		
Gly	Tyr	Lys	Val	Pro	Lys	Asn	Pro	Glu	Leu	Gly	Ser	Asp	Ala	Ser	Arg
		755					760					765			
Val	Gln	Lys	Glu	Glu	Gln	Arg	Lys	Ile	Asp	Glu	Ala	Glu	Pro	Leu	Ser
	770					775					780				
Glu	Glu	Glu	Leu	Ala	Glu	Lys	Glu	Lys	Leu	Leu	Thr	Gln	Gly	Phe	Thr
	785				790					795					800
Asn	Trp	Thr	Lys	Arg	Asp	Phe	Asn	Gln	Phe	Ile	Lys	Ala	Asn	Glu	Lys
				805					810					815	
Tyr	Gly	Arg	Asp	Asp	Ile	Asp	Asn	Ile	Ser	Lys	Glu	Val	Glu	Gly	Lys
			820					825					830		
Thr	Pro	Glu	Glu	Val	Arg	Ala	Tyr	Ser	Glu	Val	Phe	Trp	Glu	Arg	Cys
		835					840					845			
Asn	Glu	Leu	Gln	Asp	Ile	Asp	Arg	Ile	Met	Gly	Gln	Ile	Asp	Arg	Gly
	850					855					860				
Glu	Ala	Lys	Ile	Gln	Arg	Arg	Ala	Ser	Ile	Lys	Lys	Ala	Leu	Asp	Thr
	865				870					875					880
Lys	Met	Ser	Arg	Tyr	Arg	Ala	Pro	Phe	His	Gln	Leu	Arg	Ile	Ser	Tyr
				885					890					895	
Gly	Thr	Asn	Lys	Gly	Lys	Asn	Tyr	Thr	Glu	Glu	Glu	Asp	Arg	Phe	Leu
		900						905					910		
Val	Cys	Met	Leu	His	Lys	Leu	Gly	Phe	Asp	Lys	Glu	Asn	Val	Tyr	Glu
		915					920					925			
Glu	Leu	Arg	Ala	Met	Val	Arg	Cys	Ala	Pro	Gln	Phe	Arg	Phe	Asp	Trp
	930					935					940				
Phe	Ile	Lys	Ser	Arg	Thr	Ala	Met	Glu	Leu	Gln	Arg	Arg	Cys	Asn	Thr
	945				950					955					960
Leu	Ile	Thr	Leu	Ile	Glu	Arg	Glu	Asn	Gln	Glu	Leu	Glu	Glu	Arg	Glu
			965					970						975	
Arg	Ala	Glu	Lys	Arg	Lys	Gly	Arg	Gly	Ser	Gly	Arg	Gly	Pro	Gly	Ser
			980					985					990		
Gly	Lys	Arg	Lys	Gly	Asp	Gly	Ser	Ile	Ser	Ser	Pro	Pro	Pro	Val	Pro

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995	1000	1005	
Gly Gln Gly Asp Lys Asn Ser Pro Ala Arg Lys Lys Lys Lys Met			
1010	1015	1020	
 <210> SEQ ID NO 12			
<211> LENGTH: 1316			
<212> TYPE: DNA			
<213> ORGANISM: <i>Euchistus heros</i>			
 <400> SEQUENCE: 12			
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ggctcccact gagtctgtct atcagtttga aggcgaagat tatagagaaa agcaaaaagt	120		
ttttggaatt ggaaattgga ttgaaccacc aaaacgagaa cgtaaagcaa attatgcagt	180		
agatgcctat tttagagaag cactgagagt ttcagaacct aaagctccaa aggccctag	240		
gccaccaaag caaccatag ttcaagattt ccaatttttc ccacctcgtc tgtttgagct	300		
gttagatcaa gaaatatact attttcgaaa aactgtttgc tacaaggttc ctaaaaatcc	360		
ggagttagga tcagatgctt ctctgtatca aagggaagag caaagaaaaa ttgatgaagc	420		
tgagccgttg actgaggaag agctagctga gaaagaaaac ttattgacct agggttttac	480		
taattggact aaaagagatt ttaaccagtt cataaaagct aatgaaaaat atggacgtga	540		
tgatattgat aatatctcaa aagatgttga aggggaagact ccagaagaag tacgagcata	600		
ctctgaagta ttttgggaaa ggtgcaatga actacaggcc atagatcgta tcatggggca	660		
gattgataga ggtgaagcga aaattcaaag aagagccagt attaaaaag ctttagatac	720		
aaagatgagt cgatatagag caccgtttca tcaactacga attgcttatg gtacgaacaa	780		
ggggaaaaat tacacagaag aagaagacag attccttgtg tgcattgtac ataagcttgg	840		
ctttgataaa gaaaatgtgt atgaggaact tagggcgatg gtgaggtgtg ctctcagtt	900		
taggtttgat tggttcatca agtctcgaa agctttggaa ttgcaaagac gttgtaatac	960		
tctaatacag ttaattgaaa gggaaaacca agaattagaa gaaagggaaa aagtagaaaa	1020		
aaggaaaagt cgaggcagta atgggcgtgg tcccagttct ggtaaacgta agggagatgg	1080		
atctatttca tctccacctg tctctgtaca gagtataaaa agcagccctg ctcggaaaaa	1140		
gaaaaagtat atctctgttg agtaaattta tcttaaaact gggagtagat acccaattct	1200		
cattatcggtg tgatcaagga atcaatctca tataggagcc taaaacttca ttagtttgta	1260		
attgaatatt taatttacat ctctagtctt caaatattgt ttcttttaca tctgta	1316		
 <210> SEQ ID NO 13			
<211> LENGTH: 387			
<212> TYPE: PRT			
<213> ORGANISM: <i>Euchistus heros</i>			
 <400> SEQUENCE: 13			
Met Asn Lys Lys Leu Glu Gln Leu Gly Val Asp Ser Ser Leu Lys Asp			
1	5	10	15
Phe Met Met Glu Ala Pro Thr Glu Ser Val Tyr Gln Phe Glu Gly Glu			
20	25	30	
Asp Tyr Arg Glu Lys Gln Lys Val Phe Gly Ile Gly Asn Trp Ile Glu			
35	40	45	
Pro Pro Lys Arg Glu Arg Lys Ala Asn Tyr Ala Val Asp Ala Tyr Phe			
50	55	60	
Arg Glu Ala Leu Arg Val Ser Glu Pro Lys Ala Pro Lys Ala Pro Arg			
65	70	75	80

[illegible]

<400> SEQUENCE: 14

gataaatatg aataagaaaa ttttaaattt atttgtttca ttaaaaaatt atcttatggg	60
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aacaaaccca atctctagta tgcgtcctct gctgttcttg ttcactctga gtttctttat	180
cttcacaaa agcaaaactt gcaactttaa aagcagaaag taattcatca ccaacagtgg	240
ctggctcttc atctctgggt tcagctcttc tcaaaatttc gtcaatgtca caagttgggt	300

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cttcacacc atcttcttca tctttaaata attcttcagc cccaaatfff aaaatagcag 360
taagttcttc tttgttaaaa ggcgcactgg atgaagaatt ttttttatcc aggacagtcc 420
tacctgtagt atccattott tgtataacta aatgatctaa gaccattfff tgtttggccc 480
gctcgacaat attttcttca acagaacttt tagtaacaag tctgtatatg ttcacctgat 540
ttttctgacc gattctatga gctctagctt gtgcttgcaa atcattttgt ggattccaat 600
cagagtcaaa tataatgaca gtatcagctg ttgctaaatt aatgcccaaa ccaccagcac 660
gagttgataa taagaaacag aaatctggtg aattttcagc attgaaatga tcgagggcct 720
gttttctcaa ttcaccttta attgaaccgt ctaaacgttg gaaagggaaa tgtctcattt 780
gaagatactc agccagtata tccaacattc gtaccatttg agaaaatata agtactctat 840
gccagtttc ttttaaggoga acaagcaact tgtccaacag aagtaatttc cctgagcctt 900
ttaacaattg ctgtaagtag tcttcagttt ttgcttcatt ttctaattgt tttattagat 960
gtgcatgatt acagcatttt ttaattcaa taacaatatt tataaatgta ctaggagaac 1020
ctttgactcc ttttcgaaga gcagaataat ttttggaaca aatccacctg taatactgct 1080
tctgtacaga tgtcatttca acacgtaata tttgttcac tttagctggt aaagatttct 1140
caacatcctt cttaactcgt cgtagaatat atgggtccag ctgtctgtgc aacttagtat 1200
agcctttatt agcagagttg tcatgttctt tttcaaattc tcccagttta ttaaactctg 1260
tgggcataat aaagtgaagc aacgcccaaa gctctttaag actattttgc aaaggagtgc 1320
ctgttataag aagcctatgg ttggtatcaa actctttcaa tgttttgtat aataatgaat 1380
catcattttt caatctgtgt gcttcatcaa ccataaggat agcccagctt atactacca 1440
aaaatgcttt gtctttaaga acaatttcat atgtagtaag aatggcattg aattttaacc 1500
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caatataagt tacaacattc atttctggag ccataatga aaactccctc tgccatgaag 1620
tcatcgtaga taaagggaca acaattaaaa atggtccata caactggtga gtatgaaata 1680
aataatacaa actgcagata gtctgaatag ttttaccag acccatttca tcagccaaaa 1740
taatagaatt ttctttacac cacgaatgaa ccaaccaatt caaaccactg atttgataat 1800
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<210> SEQ ID NO 15

<211> LENGTH: 1454

<212> TYPE: PRT

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 15

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Met Pro Gln Lys Asp Gly Ser Glu Asp Ser Ala Ser Glu Ser Asp Lys
1           5           10           15

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Asp Gln Gly Asn Gln Glu Glu Ser Asp Asn Ser Ser Ser Glu Ser Gly
20           25           30

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Ser Gly Ser Glu Ser Asp Ser Ala Ser Ser Ala Ser Ser Ser Ser Lys
35           40           45

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Ser Ser Asp Ser Gly Ser Asp Tyr Lys Ser Lys Thr Ser Asn Ser Ser
50           55           60

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Arg Gly Lys Asn Asp Ile Lys Gln Tyr Trp Glu Asn Pro Asp Val
65           70           75           80

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Tyr Gly Ile Arg Arg Ser Asn Arg Gln Arg Lys Glu Pro Ser Arg Leu
85           90           95

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Asn Thr Gly Asp Ser Asp Ser Ser Glu Lys Thr Lys Arg Ser Val Lys

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100							105					110				
Arg	Ser	Ser	Pro	Lys	Ser	Trp	Asn	Ser	Asp	Thr	Ser	Tyr	Asp	Ser	Glu	
		115					120					125				
Thr	Asp	Lys	Glu	Ser	Lys	Arg	Pro	Pro	Pro	Ser	Lys	Pro	Pro	Gly	Gly	
	130					135					140					
Arg	Arg	Arg	Pro	Ala	Lys	Thr	Thr	Arg	Lys	Pro	Lys	Ser	Arg	Ile	Arg	
	145				150					155					160	
Asn	Arg	Ala	Tyr	Ser	Asp	Ser	Ser	Glu	Ser	Ser	Tyr	Glu	Ser	Glu	Asp	
				165					170					175		
Asp	Asn	Asn	Arg	Arg	Thr	Lys	Ser	Arg	Arg	Gly	Val	Thr	Ser	Val	Ser	
			180					185					190			
Tyr	Lys	Glu	Ala	Ser	Asp	Glu	Lys	Thr	Asp	Ser	Asp	Glu	Leu	Leu	Glu	
		195				200						205				
Pro	Asp	Pro	Glu	Pro	Val	Glu	Pro	Ala	Pro	Pro	Asp	Thr	Ser	Glu	Thr	
	210					215					220					
Ile	Glu	Lys	Val	Leu	Ala	Gln	Arg	Ile	Gly	Lys	Lys	Gly	Val	Val	Gly	
	225				230					235					240	
Asn	Gln	Thr	Thr	Val	Tyr	Ala	Val	Glu	Glu	Asn	Gly	Asp	Pro	Asn	Ser	
				245					250					255		
Asn	Tyr	Glu	Ser	Leu	Asp	Lys	Asp	Glu	Thr	Glu	Val	Gln	Tyr	Leu	Ile	
			260					265					270			
Lys	Trp	Lys	Gly	Trp	Ser	His	Ile	His	Asn	Thr	Trp	Glu	Ser	Glu	Leu	
		275				280						285				
Ser	Leu	Lys	Glu	Gln	Lys	Val	Lys	Gly	Val	Lys	Lys	Leu	Glu	Asn	Phe	
	290					295					300					
Val	Lys	Arg	Glu	Glu	Asp	Ile	Arg	Phe	Trp	Lys	Glu	His	Thr	Thr	Pro	
	305				310					315					320	
Glu	Asp	Ile	Glu	Tyr	Tyr	Glu	Cys	Gln	Leu	Glu	Leu	Gln	Gln	Glu	Leu	
			325						330					335		
Leu	Lys	Ser	Tyr	Asn	Arg	Val	Glu	Arg	Ile	Ile	Ala	Val	Ser	Lys	Thr	
			340					345					350			
Asp	Gly	Gln	Val	Glu	Tyr	Tyr	Val	Lys	Trp	Glu	Ser	Leu	Pro	Tyr	Ser	
		355					360					365				
Glu	Ala	Thr	Trp	Glu	Asp	Ser	Gly	Leu	Ile	Glu	Lys	Lys	Trp	Pro	Lys	
	370					375					380					
Lys	Ile	Lys	Glu	Phe	Lys	Glu	Arg	Glu	Asp	Ser	Lys	Arg	Thr	Pro	Ser	
	385				390					395					400	
Lys	Leu	Cys	Arg	Val	Leu	Lys	Ala	Arg	Pro	Lys	Phe	Ile	Lys	Ile	Glu	
				405					410					415		
Asp	Gln	Pro	Glu	Tyr	Met	Gly	Gly	Asp	Gln	Val	Leu	Val	Leu	Arg	Asp	
			420					425					430			
Tyr	Gln	Ile	Ser	Gly	Leu	Asn	Trp	Leu	Val	His	Ser	Trp	Cys	Lys	Glu	
		435				440						445				
Asn	Ser	Ile	Ile	Leu	Ala	Asp	Glu	Met	Gly	Leu	Gly	Lys	Thr	Ile	Gln	
	450					455					460					
Thr	Ile	Cys	Ser	Leu	Tyr	Tyr	Leu	Phe	His	Thr	His	Gln	Leu	Tyr	Gly	
	465				470					475					480	
Pro	Phe	Leu	Ile	Val	Val	Pro	Leu	Ser	Thr	Met	Thr	Ser	Trp	Gln	Arg	
				485					490					495		
Glu	Phe	Ser	Leu	Trp	Ala	Pro	Glu	Met	Asn	Val	Val	Thr	Tyr	Ile	Gly	
			500					505					510			
Asp	Ile	Asn	Ser	Arg	Asp	Val	Ile	Arg	Asn	Tyr	Glu	Trp	Cys	Tyr	Ser	
		515					520					525				

Gly 530	Ser	Lys	Arg	Leu	Lys	Phe 535	Asn	Ala	Ile	Leu	Thr 540	Thr	Tyr	Glu	Ile
Val 545	Leu	Lys	Asp	Lys	Ala 550	Phe	Leu	Gly	Ser	Ile 555	Ser	Trp	Ala	Ile	Leu 560
Met	Val	Asp	Glu	Ala 565	His	Arg	Leu	Lys	Asn 570	Asp	Asp	Ser	Leu	Leu	Tyr 575
Lys	Thr	Leu	Lys	Glu 580	Phe	Asp	Thr	Asn 585	His	Arg	Leu	Leu	Ile 590	Thr	Gly
Thr	Pro	Leu	Gln	Asn 595	Ser	Leu	Lys	Glu 600	Leu	Trp	Ala	Leu 605	Leu	His	Phe
Ile 610	Met	Pro	Asn	Arg	Phe 615	Asn	Asn	Trp	Glu	Glu	Phe 620	Glu	Lys	Glu	His
Asp 625	Asn	Ser	Ala	Asn 630	Lys	Gly	Tyr	Thr	Lys	Leu 635	His	Arg	Gln	Leu	Glu 640
Pro	Tyr	Ile	Leu	Arg 645	Arg	Val	Lys	Lys	Asp 650	Val	Glu	Lys	Ser	Leu	Pro 655
Ala	Lys	Val	Glu	Gln 660	Ile	Leu	Arg	Val 665	Glu	Met	Thr	Ser	Val 670	Gln	Lys
Gln	Tyr	Tyr	Arg	Trp 675	Ile	Leu	Ser	Lys 680	Asn	Tyr	Ser	Ala 685	Leu	Arg	Lys
Gly 690	Val	Lys	Gly	Ser	Pro 695	Ser	Thr	Phe	Ile	Asn	Ile 700	Val	Ile	Glu	Leu
Lys 705	Lys	Cys	Cys	Asn 710	His	Ala	His	Leu	Ile	Lys 715	Pro	Leu	Glu	Asn	Glu 720
Ala	Lys	Thr	Glu	Asp 725	Tyr	Leu	Gln	Gln	Leu 730	Leu	Lys	Gly	Ser	Gly	Lys 735
Leu	Leu	Leu	Leu	Asp 740	Lys	Leu	Leu	Val 745	Arg	Leu	Lys	Glu	Thr 750	Gly	His
Arg	Val	Leu	Ile	Phe 755	Ser	Gln	Met	Val 760	Arg	Met	Leu	Asp 765	Ile	Leu	Ala
Glu 770	Tyr	Leu	Gln	Met	Arg 775	His	Phe	Pro	Phe	Gln	Arg 780	Leu	Asp	Gly	Ser
Ile 785	Lys	Gly	Glu	Leu	Arg 790	Lys	Gln	Ala	Leu	Asp 795	His	Phe	Asn	Ala	Glu 800
Asn	Ser	Pro	Asp	Phe 805	Cys	Phe	Leu	Leu	Ser 810	Thr	Arg	Ala	Gly	Gly	Leu 815
Gly	Ile	Asn	Leu	Ala 820	Thr	Ala	Asp	Thr 825	Val	Ile	Ile	Phe 830	Asp	Ser	Asp
Trp	Asn	Pro	Gln	Asn 835	Asp	Leu	Gln	Ala 840	Gln	Ala	Arg	Ala 845	His	Arg	Ile
Gly 850	Gln	Lys	Asn	Gln	Val 855	Asn	Ile	Tyr	Arg	Leu	Val 860	Thr	Lys	Ser	Ser
Val 865	Glu	Glu	Asn	Ile 870	Val	Glu	Arg	Ala	Lys	Gln 875	Lys	Met	Val	Leu	Asp 880
His	Leu	Val	Ile	Gln 885	Arg	Met	Asp	Thr 890	Thr	Gly	Arg	Thr	Val	Leu	Asp 895
Lys	Lys	Asn	Ser	Ser 900	Ser	Ser	Ala	Pro 905	Phe	Asn	Lys	Glu 910	Glu	Leu	Thr
Ala	Ile	Leu	Lys	Phe 915	Gly	Ala	Glu	Glu 920	Leu	Phe	Lys	Asp 925	Glu	Glu	Asp
Gly 930	Asp	Glu	Glu	Pro 935	Thr	Cys	Asp	Ile 940	Asp	Glu	Ile 940	Leu	Arg	Arg	Ala

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Glu Thr Arg Asp	Glu Gly Pro Ala Thr Val Gly Asp	Glu Leu Leu Ser
945	950	955 960
Ala Phe Lys Val	Ala Ser Phe Ala Phe Asp	Glu Asp Lys Glu Thr Gln
965	970	975
Ser Glu Pro	Glu Gln Gln Asp Asp Thr Arg Asp	Trp Asp Glu Ile
980	985	990
Ile Pro Glu Thr Tyr Arg	Gln Lys Val Glu Glu Glu Glu Arg Ala Lys	
995	1000	1005
Glu Met Glu Asp	Leu Tyr Leu Pro Pro Arg Ser Arg	Lys Thr Leu
1010	1015	1020
Gln Gln Ile Asn His Ser	Glu Ser Asp Ala Asp Gly	Lys Ala Asn
1025	1030	1035
Lys Lys Lys Arg Lys Lys	Gly Glu Glu Asn Glu Thr Thr Glu Glu	
1040	1045	1050
Gly Ser Asp Glu Glu Lys	Pro Arg Lys Arg Gly Arg	Pro Arg Gly
1055	1060	1065
Asn Lys Gly Ser Ser Lys	Glu Val Ile Lys Gly Phe Asn Asp Ala	
1070	1075	1080
Glu Ile Arg Arg Phe Ile Arg	Ser Phe Lys Lys Phe Pro Ala Pro	
1085	1090	1095
Leu Lys Arg Leu Asp Ala Ile	Ala Cys Asp Ala Glu Leu Gln Glu	
1100	1105	1110
Lys Pro Leu Ala Glu Leu Arg	Lys Leu Gly Asp Met Leu Lys Gln	
1115	1120	1125
Arg Cys Lys Ala Cys Leu Gly	Asp Gln Thr Lys Glu Asn Leu Thr	
1130	1135	1140
Asp Ala Asn Glu Glu Asn Thr	Gly Thr Ser Gly Arg Lys Arg Gly	
1145	1150	1155
Arg Gly Pro Ser Ala Lys Leu	Gly Gly Val Ser Val Asn Ala Lys	
1160	1165	1170
Ser Leu Leu Ala Cys Glu Lys	Glu Leu Glu Pro Leu Asp Ile Glu	
1175	1180	1185
Ile Pro Leu Asp Pro Asn Glu	Arg Asn Lys Trp Val Leu Asp Val	
1190	1195	1200
Arg Val Lys Pro Ala Asn Phe	Asp Cys Asp Trp Asp Val Asn Asp	
1205	1210	1215
Asp Ser Ala Leu Leu Arg Gly	Val Tyr Gln Tyr Gly Met Gly Ser	
1220	1225	1230
Trp Glu Ala Ile Lys Met Asp	Pro Ser Ile Gly Ile Ser Asp Lys	
1235	1240	1245
Ile Leu Ser Asn Asn Gly Ser	Lys Pro Gln Thr Lys His Leu Ala	
1250	1255	1260
Ser Arg Ala Glu Tyr Leu Leu	Lys Val Leu Lys Lys Ser Ile Asp	
1265	1270	1275
Gln Arg Gln Gly Ser Thr Val	Lys Thr Lys Arg Gln Arg Lys Arg	
1280	1285	1290
Asp Asn Lys Ala Thr Ser Arg	Glu Ile Ile Glu Asp Lys Asp Asp	
1295	1300	1305
Ser Ser Gly Gly Glu Leu Pro	Ala Glu Ser Val Ser Thr Pro Ser	
1310	1315	1320
Gln Asp Ser Phe Asn His Lys	Asp Ile Lys Leu Glu Glu Asn Glu	
1325	1330	1335
Glu Asp Lys Lys Lys Gly Lys	Lys Lys Glu Thr Gln Lys Lys Lys	

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1340	1345	1350
Lys Lys Asn Glu Ser Gly Pro Met His Phe Thr Ala Asn Ser Glu		
1355	1360	1365
Pro Arg Ala Leu Asp Val Leu Gly Asp Leu Glu Pro Ser Ile Phe		
1370	1375	1380
Asn Glu Cys Lys Glu Lys Met Arg Pro Val Lys Lys Ala Leu Lys		
1385	1390	1395
Ala Leu Asp Asn Pro Asp Gln Ser Leu Gly Pro Gln Glu Gln Val		
1400	1405	1410
Asn His Thr Arg Gln Cys Leu Val Gln Ile Gly Asp Gln Ile Asn		
1415	1420	1425
Lys Cys Leu Met Glu Tyr Lys Glu Ser Asp Ile Ile Lys Gln Trp		
1430	1435	1440
Arg Arg Cys Val Ser Ser Asn Phe Val Ile Val		
1445	1450	

<210> SEQ ID NO 16
 <211> LENGTH: 496
 <212> TYPE: DNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 16

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ttgtccactc gtgcgggagg tcttggtatt aatctcgcta ctgcagatac agttattatt      180
tatgactctg actggaatcc tcataacgat attcaggcct ttctgagagc acacaggata      240
gggcaagcaa acaagggtat gatttatcga tttgtgacac gagcgtctgt tgaagaaaga      300
gtaacgcaag tggctaagag aaaaatgatg ttaaccatc ttgtcgtacg accaggtatg      360
gggtggcaagc aagcaatttt cactaagcaa gaacttgatg atattttaag gtttgaaca      420
gaagaacttt tcaaagaaga gcagggtaaa gaagatgaag ccattcatta tgacgataaa      480
gctgttgaag aattac                                     496

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<210> SEQ ID NO 17
 <211> LENGTH: 481
 <212> TYPE: DNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 17

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gtcctagaaa ggaaactaat aaattggcac cagccggtgg tgatggttct gccgaccatc      180
ggcatcgtat gaccgagcag gaagaagatg aagaactgct tgctgaaagt aatacttctt      240
caaaatcctt agcaaggttt gacgcttctc ctttttatat taaaagcgga gagttgaggg      300
attaccagat acgtggtttg aattggatga tatccctcta cgaacacggt ataaatggta      360
tacttgctga tgagatgggt ttaggtaaaa ctctccaaac tatttctctc cttggttaca      420
tgaagcatta tagaaatata ccagggccac atatggtcat cgtacccaaa tcaacattag      480
c                                     481

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<210> SEQ ID NO 18
 <211> LENGTH: 490
 <212> TYPE: DNA

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<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 18

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tattttcgaa aaactgtttg ctacaagggt cctaaaaatc cggaggttagg atcagatgct	120
tctcgtatatac aaaggggaaga gcaaagaaaa attgatgaag ctgagccgtt gactgaggaa	180
gagctagctg agaaagaaaa cttattgacc cagggtttta ctaattggac taaaagagat	240
tttaaccagt tcataaaagc taatgaaaaa tatggacgtg atgatattga taatatctca	300
aaagatgttg aagggaagac tccagaagaa gtacgagcat actctgaagt attttgggaa	360
aggtgcaatg aactacaggc catagatcgt atcatggggc agattgatag aggtgaagcg	420
aaaattcaaa gaagagccag tattaaaaaa gctttagata caaagatgag tcgatataga	480
gcaccgtttc	490

<210> SEQ ID NO 19

<211> LENGTH: 496

<212> TYPE: DNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 19

cagctggaac catatattct acgacgagtt aagaaggatg ttgagaaatc ttaccagct	60
aaagtggaac aaatattacg tgttgaaatg acatctgtac agaagcagta ttacaggtagg	120
attttgtcca aaaattattc tgccttcga aaaggagtca aaggttctcc tagtacattt	180
ataaatattg ttattgaatt aaaaaaatgc tgtaatcatg cacatctaataaaaaccatta	240
gaaaatgaag caaaaactga agactactta cagcaattgt taaaaggctc agggaaatta	300
cttctgttgg acaagttgct tgttcgcctt aaagaaactg ggcataagag acttatattt	360
tctcaaatgg tacgaatggt ggataactg gctgagtac ttcaaatgag acatttcct	420
ttccaacggt tagacggttc aattaaaggt gaattgagaa agcaagccct cgatcatttc	480
aatgctgaaa attcac	496

<210> SEQ ID NO 20

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer Mi2.T7.F

<400> SEQUENCE: 20

taatacgact cactataggg aagaaggcat agaacaga	38
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<210> SEQ ID NO 21

<211> LENGTH: 39

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer Mi2.T7.R

<400> SEQUENCE: 21

taatacgact cactataggg tcagaatggt aatcagaga	39
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<210> SEQ ID NO 22

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer ISWI30.T7.F

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<400> SEQUENCE: 22

taatacgact cactataggg tgaatcagtc taccaatt 38

<210> SEQ ID NO 23

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer ISWI30.T7.R

<400> SEQUENCE: 23

taatacgact cactataggg ggttctgact catctatt 38

<210> SEQ ID NO 24

<211> LENGTH: 39

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer ISWI2.T7.F

<400> SEQUENCE: 24

taatacgact cactataggg ttgctcaatc ctacataca 39

<210> SEQ ID NO 25

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer ISWI2.T7.R

<400> SEQUENCE: 25

taatacgact cactataggg gaataccaac aggctact 38

<210> SEQ ID NO 26

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer KSMT.T7.F

<400> SEQUENCE: 26

taatacgact cactataggg gatcaaattc aagcaact 38

<210> SEQ ID NO 27

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer KSMT.T7.R

<400> SEQUENCE: 27

taatacgact cactataggg ttcttcctaa accatgtt 38

<210> SEQ ID NO 28

<211> LENGTH: 38

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer CHD1.T7.F

<400> SEQUENCE: 28

taatacgact cactataggg ttgtcttctt tctttcaa 38

<210> SEQ ID NO 29

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<211> LENGTH: 39
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Primer CHD1.T7.R

<400> SEQUENCE: 29

taatacgact cactataggg cttctttgtt aaacggatt 39

<210> SEQ ID NO 30
 <211> LENGTH: 4493
 <212> TYPE: DNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 30

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 actaggaaga attctcgacc tttatttgtg gctggtggtt ttgcagcggc agctgagaag 120
 atgcctgagt aggctgatt gagagtgcag gtacggatgg ctttctctcc ggctctctca 180
 gtcctctggg gccacgtgag ccaatatgac cagggccgac gggcctcaac cgtggcctcc 240
 ctggacctcg acgattgctt cggttgctta aagcaggcat gaaaggcctc gaaatttttg 300
 gtggtgtctg gctgataggt tcctctggtt cgggtgtgtt atcattggag tcttggtctg 360
 atttcataac gtcctgccc ttcggactct cagctggtga ttccacacct tggtaaaactt 420
 caacgacccc atcttcagtc ttcatatgtt tcaccgtgct cttgtaattt cctagagcat 480
 tttttctttt tctttctctt tcctgtaga tatcttgctt tgattcttca agttgtctcc 540
 tctctgcttg cttctgcoga tatttctgtt ctatttctgc atcatcaagc aatagagaaa 600
 caacctcttt cggtttaaga gtgtctggtt tgaaattacc cccactgatg acaacccttt 660
 ggatttcaact tttttcttga gctctctgca agatccttcc ttcgatggtc cctttacaaa 720
 tgagccgata gactgtgact tgtttcgttt gtccaagacg atgggcacgg tccatcgctt 780
 gttggtcaac agtaggggtc caatcactgt catagaatat cacagtatca gctgcggtaa 840
 ggttgattcc aagacctcca gctcgcgtac tcaacaggaa tacaatatg tcctctcttg 900
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 tatatgtgtg ctttctgtac cacatatatt cctctaacia gtcatcatc cttgtcatct 1020
 gggagtaaat tagagctcga tgcccttgtt ccttcagcct ggtgagaagc ccatccaaga 1080
 cgtacagctt tccagcatca gttaccagtg tctgtttgtc aggtatgact atactcgacc 1140
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 aagagatcca gctccacctt cggttcagac tgtacagccg tctgcttttc tttatctcca 1320
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aagaagatcc	agaagagtgt	agcaaatctt	caattcgaat	cttctttttc	acagctgaat	2040
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aaaatattac taaactgaaa tgtaaaactt atatagaatc atatttaaaa tgagttgaac 4260
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gtcaaataaa aaaggaaagt gaaattaggt tagtatatat tgaaagacgc atctcctttt 4440
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<210> SEQ ID NO 31
<211> LENGTH: 1363
<212> TYPE: PRT
<213> ORGANISM: Euchistus heros

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<400> SEQUENCE: 31

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Met Ser Asp Asp Lys Thr Asn Met Val Val Lys Thr Glu Ile Ala Lys
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Pro Leu Tyr Tyr Gln Lys Leu Glu Lys Ser Leu Asp Ile Ala Pro Phe
20          25          30
Leu Thr Tyr Val Glu Asp Phe Leu Lys Gln Pro Leu Gly Glu Asp Gly
35          40          45
Glu Ala Ser Ser Asp Ser Asp Thr Glu Leu Ala Gly Val Asp Ala Tyr
50          55          60
Thr Gly Glu Thr Val Trp Asn Gly Ile Thr Ala Thr Lys Glu Asp Arg
65          70          75          80
Ile Ala Asp Lys Lys Arg Leu Tyr Asn Leu Ser Asn Val Ser Leu Asp
85          90          95
Arg Gln Trp Leu Val Asp Val Leu Leu Thr Asp Thr Ser Asp Ser Ser
100         105         110
Asp Asp Glu Glu Ile Thr Glu Glu Asp Leu Gln Asp Met Leu Arg Glu
115         120         125
His Val Leu Arg Asn Lys Tyr Lys Lys Lys Phe Tyr His Asn Ser Lys
130         135         140
Asn Gln Glu Tyr Met Tyr Tyr Gly Thr Gly Leu Leu Ser Asn Phe Asp
145         150         155         160
Lys Tyr Pro Glu His Gln Ser Ser Ser Ser Gly Phe Val Lys Lys Lys
165         170         175
Lys Phe Val Phe Gln Lys Lys Glu Lys Leu Lys Lys Gln Asp Gln Pro
180         185         190
His Glu Ser Phe Ser Val Glu Lys Lys Lys Arg Lys Ile Lys Glu Glu
195         200         205
Pro Asp Leu Gln Ile Met Ser Arg Ser Lys Lys Glu Met Ser Leu Arg
210         215         220
Lys Lys Ser Ala Gln Asn Lys Ala Ala Glu Ile Met Ala His Arg Arg
225         230         235         240
Arg Lys Ile Trp Ala Asn Met Ala Lys Lys Glu Ile Gly Lys Val Gln
245         250         255
Arg Tyr Arg Ile Ser Asn His Lys Glu Ile Leu Thr Ala Cys Arg Arg
260         265         270
Ala Ala Thr Asn Cys Met Arg His Cys Arg Gln Arg Ala Met Gln Ser
275         280         285
Gln Lys Asn Met Lys Glu Val Ile Trp Arg Ala Lys Arg Leu Thr Arg
290         295         300
Glu Met Gln Val Tyr Trp Lys Arg Phe Glu Arg Val Glu Arg Glu Thr
305         310         315         320
Arg Arg Arg Met Glu Lys Glu Ala Glu Glu Gln Arg Lys Leu Asp Val

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325								330					335				
Glu	Leu	Met	Glu		Ala	Lys	Arg	Gln	Gln	Arg	Lys	Leu	Asn	Phe	Leu	Ile	
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Thr	Gln	Thr	Glu	Leu	Tyr	Ala		His	Phe	Met	Ser	Arg	Lys	Leu	Gly	Gly	
			355					360					365				
Gly	Thr	Thr	Glu	Asp	Gln	Leu	Arg	Ile	Leu	Ser	Gln	Leu	Glu	Glu	Glu		
			370				375				380						
Ala	Asn	Pro	Arg	Leu	Leu	Pro	Leu	Asp	Asp	Tyr	Asp	Cys	Glu	Ala	Glu		
385					390					395						400	
Lys	Glu	Lys	Val	Arg	Lys	Lys	Val	Glu	Lys	Val	Phe	Phe	Leu	Glu	Glu		
			405						410					415			
Ala	Arg	Ala	Arg	Glu	Phe	Gly	Val	Ser	Gln	Lys	Met	Glu	Glu	Asp	Gly		
			420					425					430				
Asp	Val	Gly	Glu	Asp	Lys	Pro	Gln	Pro	Gln	Ile	Phe	Arg	Gly	Thr	Leu		
			435				440					445					
Lys	Gly	Tyr	Gln	Ile	Lys	Gly	Met	Thr	Trp	Leu	Val	Asn	Leu	Tyr	Asp		
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			485						490					495			
Trp	Gly	Pro	Phe	Leu	Ile	Ile	Ser	Pro	Ala	Ser	Thr	Leu	His	Asn	Trp		
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Gln	Gln	Glu	Met	Gln	Arg	Phe	Val	Pro	Asp	Phe	Lys	Val	Val	Pro	Tyr		
			515				520					525					
Trp	Gly	Asn	Gln	Gln	Glu	Arg	Lys	Ile	Leu	Arg	Gln	Phe	Trp	Asp	Gln		
			530			535					540						
Lys	Gly	Leu	His	Thr	Lys	Asp	Ala	Ser	Phe	His	Val	Val	Ile	Thr	Ser		
545					550					555						560	
Tyr	Gln	Leu	Val	Ile	Thr	Asp	Ile	Lys	Tyr	Phe	Asn	Arg	Ile	Lys	Trp		
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Gln	Tyr	Leu	Ile	Leu	Asp	Glu	Ala	Gln	Ala	Ile	Lys	Ser	Thr	Thr	Ser		
			580					585					590				
Met	Arg	Trp	Lys	Leu	Leu	Leu	Gly	Phe	Asn	Cys	Arg	Asn	Arg	Leu	Leu		
			595				600					605					
Leu	Ser	Gly	Thr	Pro	Ile	Gln	Asn	Ser	Met	Ala	Glu	Leu	Trp	Ala	Leu		
			610			615					620						
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625					630					635						640	
Glu	Trp	Phe	Ser	Lys	Asp	Ile	Glu	Ser	His	Ala	Glu	His	Lys	Thr	Ser		
			645						650					655			
Ile	Asp	Glu	Lys	His	Leu	Ser	Arg	Leu	His	Met	Ile	Leu	Lys	Pro	Phe		
			660					665					670				
Met	Leu	Arg	Arg	Val	Lys	Thr	Asp	Val	Glu	Asn	Glu	Leu	Ser	Asp	Lys		
			675				680					685					
Ile	Glu	Ile	Met	Val	Tyr	Cys	Pro	Leu	Thr	Thr	Arg	Gln	Lys	Met	Leu		
			690			695					700						
Tyr	Ser	Ala	Val	Lys	Lys	Lys	Ile	Arg	Ile	Glu	Asp	Leu	Leu	His	Ser		
705					710					715						720	
Ser	Gly	Ser	Ser	Tyr	Gln	Ser	Ala	Gln	Ser	Ile	Thr	Ser	Asn	Leu	Met		
			725						730					735			
Asn	Leu	Val	Met	Gln	Phe	Arg	Lys	Val	Cys	Asn	His	Pro	Glu	Leu	Phe		
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Glu Arg Arg Glu Pro Arg Ser Pro Phe Ser Met Ala Thr Glu Asp Tyr	755	760	765
Ile Val Pro Ala Leu Ile Thr Asp Ala Val Phe Pro Gly Asp Lys Leu	770	775	780
His Leu Leu Met Asn Tyr Leu Ser Pro Phe Asn Pro Phe His Ser His	785	790	795
Arg Ser Leu Phe Ser Asp Asp Ser Ser Glu Arg Asp Ser Ser Phe Ser	805	810	815
Phe Thr Arg Leu Leu Arg Leu Ser Val Gly Glu Leu Tyr Lys Leu Met	820	825	830
Phe Phe Gly Ile Tyr Phe Arg Trp His Asn Ile Ile Ala Arg Arg Gln	835	840	845
Gly Val Arg Arg Lys Leu Val Gly Ile Ile Glu Asp Arg Ser Tyr Pro	850	855	860
Asn Asn Leu Val Phe Thr Thr Tyr Ser Asn Gln Val Phe Ser His Ser	865	870	875
Thr Val Arg Ile Glu Ser Met Pro Glu Thr Ile Glu His Arg Ile Val	885	890	895
Arg Ser Lys Lys Ala Lys Gly Glu Cys Gly Gly Glu Asp Ser Ala Asn	900	905	910
Leu Leu Pro Ala Leu Pro His Ile Val Arg Lys Pro Gln Ile Leu Ser	915	920	925
Cys Gln Pro Val Tyr Leu Pro Pro Phe Leu Phe Thr Pro Thr Val Lys	930	935	940
Val Glu Ile Lys Lys Ser Arg Arg Leu Tyr Ser Ser Asn Arg Arg Trp	945	950	955
Ser Trp Ile Ser Ser His Leu Trp Gly Glu Asp Gly Arg Leu Trp Gln	965	970	975
Val Leu Trp Glu Gly Asp Lys Gln Glu Pro Gly Phe Gln Cys Pro Pro	980	985	990
Pro Leu Gly Leu Leu Ser Leu Arg Pro Ile Thr Gly Trp Ser Ser Ile	995	1000	1005
Val Ile Pro Asp Lys Gln Thr Leu Val Thr Asp Ala Gly Lys Leu	1010	1015	1020
Tyr Val Leu Asp Gly Leu Leu Thr Arg Leu Lys Glu Gln Gly His	1025	1030	1035
Arg Ala Leu Ile Tyr Ser Gln Met Thr Arg Met Ile Asp Leu Leu	1040	1045	1050
Glu Glu Tyr Met Trp Tyr Arg Lys His Thr Tyr Met Arg Leu Asp	1055	1060	1065
Gly Ser Ser Lys Ile Ser Asp Arg Arg Asp Met Val Ala Asp Phe	1070	1075	1080
Gln Thr Arg Glu Asp Ile Phe Val Phe Leu Leu Ser Thr Arg Ala	1085	1090	1095
Gly Gly Leu Gly Ile Asn Leu Thr Ala Ala Asp Thr Val Ile Phe	1100	1105	1110
Tyr Asp Ser Asp Trp Asn Pro Thr Val Asp Gln Gln Ala Met Asp	1115	1120	1125
Arg Ala His Arg Leu Gly Gln Thr Lys Gln Val Thr Val Tyr Arg	1130	1135	1140
Leu Ile Cys Lys Gly Thr Ile Glu Glu Arg Ile Leu Gln Arg Ala	1145	1150	1155

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Phe	Lys	Pro	Asp	Thr	Leu	Lys	Pro	Lys	Glu	Val	Val	Ser	Leu	Leu
1175						1180						1185		
Leu	Asp	Asp	Ala	Glu	Ile	Glu	Gln	Lys	Tyr	Arg	Gln	Lys	Gln	Ala
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Glu	Arg	Arg	Gln	Leu	Glu	Glu	Ser	Arg	Gln	Asp	Ile	Tyr	Arg	Glu
1205						1210					1215			
Arg	Glu	Arg	Lys	Arg	Lys	Asn	Ala	Leu	Gly	Asn	Tyr	Lys	Asp	Thr
1220						1225					1230			
Val	Lys	His	Met	Lys	Thr	Glu	Asp	Gly	Val	Val	Glu	Val	Tyr	Gln
1235						1240					1245			
Gly	Val	Lys	Ser	Pro	Ala	Glu	Ser	Pro	Lys	Gly	Thr	Asp	Val	Met
1250						1255					1260			
Lys	Ser	Ser	Gln	Asp	Ser	Asn	Asp	Asn	Asn	Thr	Asp	Gln	Glu	Glu
1265						1270					1275			
Pro	Ile	Ser	Gln	Thr	Pro	Pro	Lys	Ile	Ser	Arg	Pro	Phe	Met	Pro
1280						1285					1290			
Ala	Leu	Ser	Asn	Arg	Ser	Asn	Arg	Arg	Gly	Pro	Gly	Arg	Pro	Arg
1295						1300					1305			
Leu	Arg	Pro	Val	Gly	Pro	Gly	His	Ile	Gly	Ser	Arg	Gly	Pro	Arg
1310						1315					1320			
Gly	Leu	Arg	Gly	Pro	Gly	Arg	Lys	Pro	Ser	Val	Pro	Ala	Leu	Ser
1325						1330					1335			
Ile	Arg	Pro	Thr	Gln	Ala	Ser	Ser	Gln	Leu	Pro	Leu	Gln	Asn	His
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Gln	Pro	Gln	Ile	Lys	Val	Glu	Asn	Ser	Ser					
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<210> SEQ ID NO 32

<211> LENGTH: 6108

<212> TYPE: DNA

<213> ORGANISM: Euchistus heros

<400> SEQUENCE: 32

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<210> SEQ ID NO 33
<211> LENGTH: 1790
<212> TYPE: PRT
<213> ORGANISM: Euchistus heros

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<400> SEQUENCE: 33

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20          25          30
Gln Gln Gly Ser Pro Ser Met Thr Arg Leu Ala Asn Ser Ser Gln Gly
35          40          45
Gly Arg Leu Gln Val Arg Ser Ser Pro Thr Ser Ile Leu Ser Asp Ser
50          55          60
Pro Met Met Thr Asn Leu Gln Pro Pro Ser Gln Gly Ser Asn Thr Asp
65          70          75          80
Val Arg Val Pro Ser Ser Ala Ser Pro Thr Lys Ala Glu Leu Ser Ser
85          90          95
Lys Phe Gln Lys Leu Thr Thr Val Lys Ser Ala Val Lys Asn Ala Ser
100         105         110
Pro Val Asp Gly Val Asn Ser Gln Glu Gln Ile Val Glu Arg Ala Lys
115         120         125
Gln Glu Ala Tyr Val Met Gln Arg Val Gly Glu Leu Gln Lys Asp Gly
130         135         140
Leu Trp Pro Glu Lys Arg Leu Pro Lys Val His Glu Leu Pro Arg Ala
145         150         155         160
Lys Ala His Trp Asp Tyr Leu Leu Glu Glu Met Ala Trp Leu Ala Ala
165         170         175
Asp Phe Ala Gln Glu Arg Lys Trp Lys Lys Ala Ala Ala Lys Lys Cys
180         185         190
Ala Arg Met Val Gln Lys His Phe Gln Glu Lys Glu Ala Leu Val Gln
195         200         205
Lys Ala Ile Lys Ala Gln Glu Met Gln Leu Lys Arg Ile Ala Ser Phe
210         215         220
Ile Ala Lys Glu Ile Lys Gln Phe Trp Ser Asn Val Glu Lys Leu Val
225         230         235         240
Glu Phe Lys Gln Gln Thr Arg Leu Glu Glu Lys Arg Lys Lys Ala Leu
245         250         255
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Leu Val Ala Glu Ser Met Asn Thr Ser Arg Pro Thr Ser Pro Arg His
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Thr	Pro	Val	Ser	Met	Asp	Glu 355	Asp	Val	Glu 360	Asp	Asp	Asp	Met	Glu	Tyr
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Lys	Glu	Glu	Gly	Asn 405	Met	Ser	Ile	Glu	Gln 410	Leu	Arg	Ala	Lys	Tyr	Gly 415
Ile	Glu	Leu	Pro	Ser	Glu	Pro	Leu	Pro	Ser 425	Asn	Gly	Asp	Pro	Ser	Ser
Gln 435	Arg	Ser	Arg	Lys	Arg	Lys	Glu 440	Ile	Leu	Asn	Glu	Glu 445	Ser	Asp	Glu
Ala 450	Ser	Glu	Glu	Glu	Asp	Phe 455	Glu	Asp	Glu	Val	Ser 460	Glu	Glu	Glu	Glu
Glu 465	Glu	Asp	Ser	Ser	Ser 470	Ser	Asp	Lys	Glu	Asp 475	Ser	Thr	Leu	Lys	Ala 480
Leu	Ile	Asn	Asp	Ser 485	His	His	Val	Glu	Glu 490	Gly	Asn	Gln	Asp	Lys	Asp 495
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Gly 515	Asn	Thr	Leu	Ser	Ser	Thr	Ser	Val	Val 520	Thr	Lys	Val	Pro	Phe	Leu
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Val 545	Thr	Met	Tyr	Asp	Arg 550	Lys	Leu	Asn	Gly	Ile 555	Leu	Ala	Asp	Glu	Met 560
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Val 595	Met	Leu	Asn	Trp	Glu	Met	Glu	Phe	Lys 600	Lys	Trp	Cys	Pro	Ala	Phe
Lys 610	Ile	Leu	Thr	Tyr	Tyr	Gly 615	Thr	Gln	Lys	Glu	Arg 620	Lys	Leu	Lys	Arg
Ser 625	Gly	Trp	Thr	Lys	Pro 630	Asn	Ala	Phe	His	Ile 635	Cys	Ile	Thr	Ser	Tyr 640
Lys	Leu	Val	Ile	Gln 645	Asp	His	Gln	Ser	Phe 650	Arg	Arg	Lys	Lys	Trp	Lys 655
Tyr	Leu	Ile	Leu	Asp 660	Glu	Ala	Gln	Asn	Ile 665	Lys	Asn	Phe	Lys	Ser	Gln
Arg	Trp	Gln	Leu	Leu	Leu	Asn 675	Phe	Gln	Thr 680	Gln	Arg	Arg	Leu	Leu	Leu 685
Thr 690	Gly	Thr	Pro	Leu	Gln 695	Asn	Asn	Leu	Met	Glu	Leu	Trp	Ser	Leu	Met 700
His	Phe	Leu	Met	Pro	Asn	Val	Phe	Glu	Ser	His	Arg	Glu	Phe	Lys	Glu

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705	710	715	720
Trp Phe Ser Asn Pro Val Thr Gly Met Ile Glu Gly Asn Ser Glu Tyr			
725		730	735
Asn Asp Thr Ile Ile Lys Arg Leu His Lys Val Leu Arg Pro Phe Leu			
740		745	750
Leu Arg Arg Leu Lys Cys Glu Val Glu Thr Gln Met Pro Lys Lys Tyr			
755		760	765
Glu His Ile Val Met Cys Arg Leu Ser Asn Arg Gln Arg Tyr Leu Tyr			
770		775	780
Asp Asp Phe Met Ser Arg Ala Lys Thr Lys Glu Thr Leu Ala Ser Gly			
785		790	795
800			
Ser Leu Leu Ser Val Ile Asn Val Leu Met Gln Leu Arg Lys Val Cys			
805		810	815
Asn His Pro Asn Leu Phe Glu Val Arg Pro Thr Val Ser Pro Phe Gln			
820		825	830
Met Asp Ser Leu Glu Tyr His Val Pro Ser Ile Val Trp Ser Ala Leu			
835		840	845
Asp Tyr Asp Pro Phe Lys His Val Asp Leu Val Phe Leu Asn Leu Arg			
850		855	860
Ile Ile Glu Phe Glu Leu Trp Leu Leu Ala Phe Val Ala His Arg Ala			
865		870	875
880			
Arg Lys Tyr Lys Val Asn Pro Asp Phe Ile Arg Thr Ile Asp Ser Ala			
885		890	895
Pro Pro Asp Leu Pro Pro Cys Pro Lys Gly Arg Val Lys Ile Asn Val			
900		905	910
Lys Lys Arg Ile Leu Pro Gln Pro Pro Gln Val Ser Ser Pro Val Arg			
915		920	925
Pro Ile Val Gln Val Ser Asn Asn His Arg Ile Ser Ser Phe Ser Thr			
930		935	940
Lys Val Gly Thr Ser Pro Leu Ile Lys Leu Gly Gln Asn Ala Ser Gln			
945		950	955
960			
Ser Val Thr Leu Arg Leu Ala Asn Pro Thr Ser Gly Gln Ile Pro Asn			
965		970	975
Tyr Leu Gln Leu Leu Gln Pro Gly Ala Met Lys Ala Ile Ser Val Ala			
980		985	990
Ser Leu Thr Thr Thr Ser Gly Gly Val Val Lys Asn Glu Gly Asn Ala			
995		1000	1005
Gly Arg Met Val Pro Gln Phe Ala Gln Leu Val Asn Thr Pro Thr			
1010		1015	1020
Gly Arg Gln Leu Val Leu Ser Pro Gln Pro Val Ile Thr Pro Gln			
1025		1030	1035
Gly Gly Ala Thr Thr Val Met Thr Thr Thr Gly Gln Arg Leu Thr			
1040		1045	1050
Val Val Ser Lys Gln Gly Ser Asn Val Ala Lys Leu Val Asn Pro			
1055		1060	1065
Thr Ser Val Thr Gln Ile Asn Ser Asn Arg Pro Ile Met Arg Val			
1070		1075	1080
Pro Pro Leu Asn Ile Thr Ser Ala Ala Thr Val Val Gln Asn Ser			
1085		1090	1095
Leu Thr Thr Val Asn Asn Ser Pro Val Ser Ile Ser Ser Glu Pro			
1100		1105	1110
Ala Gln Gly Asn Gly Ser Asn Val Asn Ser Leu Pro Ser Leu Lys			
1115		1120	1125

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Lys Val Thr Arg Gln Ser Thr	Leu Asn Ser Asn Lys	Asn Asn Lys
1130	1135	1140
Glu Glu Ser Asn Pro Asn Glu	Asp Leu Arg Asp Glu	Tyr Leu Glu
1145	1150	1155
Asn Leu Arg Lys Lys Arg Arg	Lys Glu Lys Leu Glu	Leu Ile Val
1160	1165	1170
Arg Ile Asn Glu Gln Arg Cys	Gln Ala Leu Pro Ile	Tyr Gly Ser
1175	1180	1185
Asp Leu Leu Ser Ser Leu Thr	Leu Asp Asp Ala Pro	Asp Glu Thr
1190	1195	1200
Lys Asp Cys Ile Gly Thr Val	His Cys Lys Asn Ala	Leu Ser Ser
1205	1210	1215
Asn Pro Leu Leu Phe Trp Asn	Gln Thr His Ala Leu	Thr Ala Ala
1220	1225	1230
Ile His Ser Ile Glu Asp Arg	Val Ala Glu Leu Ser	Asp Ile Phe
1235	1240	1245
Ser Arg Phe Val Leu Tyr Val	Pro Ala Val Ser Ser	Gln Pro Val
1250	1255	1260
Arg Leu His Val Pro His Pro	Pro Pro Ser Lys Met	Phe Gln Glu
1265	1270	1275
Glu Lys Arg Leu Ser Leu Val	Asp Thr Trp Leu Arg	Pro Lys Leu
1280	1285	1290
Arg Leu Leu His Ser Ile Ser	Ser Ala Met Ser Thr	Gln Phe Pro
1295	1300	1305
Asp Arg Arg Leu Ile Gln Tyr	Asp Cys Gly Lys Leu	Gln Ser Leu
1310	1315	1320
Asp Lys Leu Leu Arg Arg Leu	Lys Ala Asp His His	Arg Ile Leu
1325	1330	1335
Ile Phe Thr Gln Met Thr Arg	Met Leu Asp Val Leu	Glu Ala Phe
1340	1345	1350
Leu Asn Phe His Gly His Ile	Tyr Leu Arg Leu Asp	Gly Thr Thr
1355	1360	1365
Lys Val Asp Gln Arg Gln Leu	Leu Met Glu Arg Phe	Asn Ala Asp
1370	1375	1380
Lys Arg Ile Phe Cys Phe Ile	Leu Ser Thr Arg Ser	Gly Gly Val
1385	1390	1395
Gly Ile Asn Leu Thr Gly Ala	Asp Thr Val Ile Phe	Tyr Asp Ser
1400	1405	1410
Asp Trp Asn Pro Thr Met Asp	Ala Gln Ala Gln Asp	Arg Cys His
1415	1420	1425
Arg Ile Gly Gln Thr Arg Asp	Val His Ile Tyr Arg	Leu Ile Ser
1430	1435	1440
Glu Lys Thr Val Glu Glu Asn	Ile Leu Lys Lys Ala	Asn Gln Lys
1445	1450	1455
Arg Met Leu Gly Asp Val Ala	Ile Glu Gly Gly Asn	Phe Thr Thr
1460	1465	1470
Ala Tyr Phe Lys Ser Ser Thr	Ile Gln Asp Leu Phe	Asn Val Asp
1475	1480	1485
Thr Ser Glu Asn Asp Ala Ser	Arg Arg Met Ala Glu	Val Leu His
1490	1495	1500
Asn Ser Asp Lys Thr Ala Gln	Asn Pro Glu Asp Thr	Gln Met Val
1505	1510	1515

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Ala Asn Ser Asp Glu Lys Val	Ala Ile Gly Ala Leu Glu Ser Ala
1520 1525	1530
Met Ala Gln Ala Glu Asp Glu	Ser Asp Val Ala Ala Ala Lys Val
1535 1540	1545
Ala Lys Ala Glu Ala Ala Ala	Glu Leu Ala Glu Phe Asp Glu Asn
1550 1555	1560
Ile Pro Ile Asp Thr His Glu	Gly Val Gly Gln Glu Met Ser Lys
1565 1570	1575
Ala Glu Leu Glu Leu His Asn	Leu Met Gln Gln Leu Ser Ala Val
1580 1585	1590
Glu Arg Tyr Ala Met Lys Phe	Met Glu Glu Asn Asp Ala Ala Trp
1595 1600	1605
Ser Lys Glu Gln Leu Ala Ala	Ala Glu Ala Glu Ile Glu Gln Gln
1610 1615	1620
Lys Lys Glu Trp Glu Ala Gly	Arg Leu Ala Ala Leu Gln Gly Asp
1625 1630	1635
Arg Lys Ser Pro Ser Gly Glu	Ala Glu Ser Glu Ala Val Leu Thr
1640 1645	1650
Tyr Ser Gly Val Asp Ser Arg	Asn Gln Val Asn Lys Thr Arg Ala
1655 1660	1665
Ser Gly Gly Gly Gly Arg Asn	Cys His Ser Gly Ser Ser Ile Glu
1670 1675	1680
Glu Gly Thr Pro Arg Thr Arg	Ser His Gly Arg Val Ser Ile Asp
1685 1690	1695
Leu Trp Thr Leu Asp Asp Ser	Pro Gln Asn Gly Gly Lys Pro Ser
1700 1705	1710
Ser Gly Gln Ser Arg Lys Phe	Lys Ser Gly Lys Arg Arg Ser Thr
1715 1720	1725
Pro Pro Pro Pro Asn Pro	Pro Pro Pro Pro Tyr Ser Asn Pro
1730 1735	1740
Asn Leu Val Ile Arg Thr Arg	Arg Ala Ser Ala Ala Ser Ser Asn
1745 1750	1755
Glu Ile Ser Lys Val Ser Lys	Lys Val Gly Arg Leu Ser Lys Thr
1760 1765	1770
Lys Arg Pro Ser Asp Val Val	Pro Leu Thr Ile Ser Thr Ser Leu
1775 1780	1785
Ala Ser	
1790	

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<210> SEQ ID NO 34
<211> LENGTH: 240
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: SNF2-helicase degenerate dsRNA sequence
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (216)..(216)
<223> OTHER INFORMATION: n is a, c, g, or t
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (222)..(222)
<223> OTHER INFORMATION: n is a, c, g, or t

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<400> SEQUENCE: 34

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cgssythctyy tmacsggyac hcctctvcar aayaarctwc chgarytstg ggcbtydcth      60

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aayttytvc tbcstcbat yttyaarwb tgytcbad tgyarcartg gttcaaygc      120

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cchtttygcha cmacbggmga raargtygar ytdaaygarg argaracvat yytkatyaty 180

mgdcgtytdc ayaargtyyt kcgwccktty ytvvtnmgdc gnytvaaaaa rgargtmgar 240

<210> SEQ ID NO 35
 <211> LENGTH: 27
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2-helicase degenerate dsRNA sequence
 <220> FEATURE:
 <221> NAME/KEY: misc_feature
 <222> LOCATION: (21)..(21)
 <223> OTHER INFORMATION: n is a, c, g, or t

<400> SEQUENCE: 35

mghgcygtbt gyythatygg ngaycar 27

<210> SEQ ID NO 36
 <211> LENGTH: 60
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2-helicase degenerate dsRNA sequence

<400> SEQUENCE: 36

tayaarctyc tvytsacmgg machccgytb caraacaayc tmgargaryt rttycatytr 60

<210> SEQ ID NO 37
 <211> LENGTH: 61
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2-helicase degenerate dsRNA sequence

<400> SEQUENCE: 37

garttygaya cbaaycaymg rctkcthath acwggbacyc ckytvcaraa ywskytdaar 60

g 61

<210> SEQ ID NO 38
 <211> LENGTH: 23
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Bromodomain degenerate dsRNA sequence

<400> SEQUENCE: 38

ytswsygaac crttyatgaa ryt 23

<210> SEQ ID NO 39
 <211> LENGTH: 65
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: HAND-SLIDE degenerate dsRNA sequence

<400> SEQUENCE: 39

gchgtvgatg cytaytymg vgargcwytv mgdgtytchg arccyaargc dccdaargch 60

cchmg 65

<210> SEQ ID NO 40
 <211> LENGTH: 36
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Chromodomain degenerate dsRNA sequence

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<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (33)..(33)
<223> OTHER INFORMATION: n is a, c, g, or t

<400> SEQUENCE: 40
mghaartrbg ayatggavga rvvdcbaar ytngar 36

<210> SEQ ID NO 41
<211> LENGTH: 56
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Chromodomain degenerate dsRNA sequence
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (29)..(29)
<223> OTHER INFORMATION: n is a, c, g, or t

<400> SEQUENCE: 41
bhggdaarad dggrkkbryb ggmaaymwna chacdrtsta ykmhrtagar gaaaay 56

<210> SEQ ID NO 42
<211> LENGTH: 471
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: YFPv2 hpRNA encoding sequence

<400> SEQUENCE: 42
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aatgttgatg gccacacott tagcatacgt gggaaaggct acggagatgc ctcaagtggga 120
aaggactagt accggttggg aaaggtatgt ttctgcttct acctttgata tatatataat 180
aattatcact aattagtagt aatatagtat ttcaagtatt tttttcaaaa taaaagaatg 240
tagtatatag ctattgcttt tctgtagttt ataagtgtgt atattttaat ttataacttt 300
tctaatatat gacaaaaaca tggtagatgt caggttgatc cgcggttact ttccactga 360
ggcatctcgg tagcctttcc caggtatgct aaaggtgtgg ccatcaacat tcccttccat 420
ctccacaacg taaggaatct tcccatgaaa gagaagtgtc ccagatgaca t 471

<210> SEQ ID NO 43
<211> LENGTH: 4958
<212> TYPE: RNA
<213> ORGANISM: Euchistus heros

<400> SEQUENCE: 43
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uggauaguua uguugauucu uuuuaaguga uuuugaagau uuccugacca uuuuauacag 120
aaaaacuauu uuaaacagcg cuauugcucc uuauaaucag ugugauucaa caacgaugga 180
cggagacagc ggugguuagg cgagcccuuc gccacagccu cagucgucac caaugccccc 240
uccacaagcu ccaucaccua ugggcccgcc gcaggcgccc ccaucgcaa ugccccuuc 300
uaaccaacag gcgccucac caaugggucc accgcaccac cccacagcc cgacagguua 360
ccaaggaggg augccacaca ugaauaggacc aaauagguguu ccuccuggua ugcagcaggc 420
uacucaaaac uuucagccuc aucagcauu gccacccac cagcaaccac caaugcagac 480
ugcuccuggu gggccugcua gugguggagg acaagaaaau cuuagcguc uccagcgugc 540
aaugauucu auggaagaga aaggguuca ggaagaacca cguuacugc agcugcuugc 600

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guugagggca	aggcaugcca	acauggaacc	uccgguuagg	ccuccaucuc	agcuuguugg	660
ggguggguuc	agcggugagg	guggugcccc	uccuccugcu	aaacacagcu	ucagcgcgaa	720
ccaacugcaa	caacuucgag	ugcagaucau	ggcguaucgc	cuacuugcua	ggaaccaacc	780
ucuuucccag	cagcuagcuu	uggcugugca	aggcaaacgc	cucgacagcc	cuggcgaguc	840
caacuaccag	cauccuccua	gugaaggagc	aggaggguuu	gguggagaag	gaaguggaga	900
cggggggaucg	ucgaacggcc	ugaugacgca	gccgaugcgu	gccccaugcc	ccccuggugg	960
ccagccccc	acggccucac	cgaugacagg	ccagauggca	ccuccuacug	ggccagcucc	1020
uguaaggcca	ccuccucccg	gugugucucc	uacaccuccg	cgcccuccuc	agcagguucc	1080
uggugcuccg	ggggccccac	aaccaagca	aaauaggguu	accaccaugc	caagaccgca	1140
ugguuuagau	cccauucua	uucuccagga	aagagagaau	agaguagccg	cuaggauugu	1200
acauagggaug	gaagaauuau	caauuuuacc	agcuacgaug	ccugaagacc	uucgaauaaa	1260
agcgcagaua	gaacuuaggg	ccuugagggu	acuuaacuuc	caaaggcaau	uaagagcaga	1320
ggugauagcu	uguacuagac	gcgauacaac	auuagaaaca	gcuguaaaug	ugaaagcuua	1380
uaaacgaacg	aagaggcaag	gcuuacggga	agccagagcu	acggaaaagc	uugaaaaaca	1440
acagaaacuu	gagacagaaa	ggaagaagag	acaaaaacac	caggaaauuc	ugagcacuau	1500
auugcaacau	ugcaaagacu	ucaaaagaau	ccauagaaau	aauguugcua	aaguugguag	1560
auuaauaag	gcugugauga	auuaccaugc	gaaugccgag	cgugaacaga	agaaagagca	1620
agaaaggaua	gaaaaagaac	guaugagaag	gcuuauggcu	gaggauagaag	aggguuacag	1680
gaaacugauu	gaucagaaaa	aagauaagag	auuggcauuc	cuucuuucac	aaacugauga	1740
auauauugcc	aaucuuacug	aaauggugaa	gcagcauaaa	auggaacaac	agcguaaagca	1800
ggaacaagaa	gagcaacaaa	aacggaagag	gaaaaagaaa	aagaagaaua	gggaaggaga	1860
uccagaugau	gaaagcucuc	agaugucaga	uuuacauguu	agcguauuag	aagcagcaac	1920
uggucggcag	cugacggggg	aggaugcucc	auuggccagc	cagcuuggga	gcugguugga	1980
ggcacacccg	ggcugggagc	cuuuggaaga	uagcgaagau	gaagaugaug	aagaggacag	2040
cgacgaggaa	ggugaugaua	acaguagauc	aaaagguggu	uuucaauga	uaggaaaaga	2100
ugaagcugau	agcaaguauu	cuguugaaga	cgaagcucga	gaaugauaa	agaaagcgaa	2160
gauugaagau	gaugaaauca	agaacacgac	cgaagaacau	acauacuaca	gcaucgcuca	2220
caccgugcau	gaaaauugca	ccgaacaagc	uucaaucaug	auuaacggua	aaugaaaaga	2280
auaucaauu	aaaggucuu	auugguuugu	uucuuuauac	aacaacaacu	ugaauggauu	2340
ccucgccgac	gagauggggc	uuggcaagac	aaucacaaca	auaggucuca	uuacuuuuu	2400
gauggagaag	aagaaaguua	augguccuua	ccucauuuu	guuccucugu	caacauuau	2460
caauuggguu	uuggaaauucg	agaaaugggc	uccuucagug	uuugugguag	cuuauaaagg	2520
uucuccugca	augaggagaa	cuuuacaau	acagaugcgc	ucgacgaagu	ucaauguccu	2580
gcucacgacc	uacgaguaug	ucaucaagga	caaggcagua	cuugcaaagu	ugcauuggaa	2640
guacaugaua	aucgacgagg	gacacaggau	gaaaaaccac	cauuguaagc	ugacgcaggu	2700
gcugaacacc	cauuuuuugg	caccucaccg	ccuccuucuc	acgggcacac	cucuccagaa	2760
caaacuaccu	gagcucuggg	cucuucuaaa	cuuucuccuc	ccguccaucu	ucaagucgug	2820
uucucaguuu	gagcaauugu	ucaaugcacc	auuugcuacc	acuggagaaa	agguugaguu	2880
gaaugaggaa	gaaacaauuu	ugaauuacag	gcguuuacau	aagguccuuc	gaccuuuccu	2940
ccuucgucga	cugaaaaagg	aagucgaaag	ucaguugcca	gagaaaaau	aaucacucgu	3000

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caagugugau augucugguc uccaacgugu acuuuauagg cacaugcaga guaaaggagu	3060
ccugcuuacc gaugguucug agaagggcaa gcaggguaaa ggaggagcua aagcgcuau	3120
gaacacgauc guccaauuga ggaagcuug caaucauccu uucauguucc aucauauuga	3180
agaaaaauau ugugaucacg ugggccagaa caacguuguc acagggccug aucuguuccg	3240
aguuucuggu aaauuugaau uccucgaucg uauauugcca aaacugaagg ccacgagcca	3300
uaggguacuu cuuucuguc aaauugacuca gcugaugacc aucauggagg auuauuuguc	3360
uuggagaggg uucuccuacc uucgucuuga ugguacgacc aaucugaag accgaggaga	3420
ucuucugaaa aaauucaaca auccagaaag ugaauuuuu auuuucuuug ucucaaccag	3480
agcuggaggu cucggauuga acuuacaggc ugcagauacu gucauuauau uugauucaga	3540
uuggaaccuu caucaggauu uacaagcuca agacagagcu cauaggauug gacagcaaaa	3600
cgaaguucgu guuuugcggc uaaugacagu aaauucuguu gaggagcgua uucuuugcgc	3660
ugcucgguac aagcugaaua uggaugagaa agucauucag gcugguauu uugaccagaa	3720
aucuacagga accgagaggg agaaaauuu gcaaaacauc cuucaucaag augaugcaga	3780
ugaugaggaa aaugaaguuc cagaugauga aaugguuauu cguaugauug cgcgaaacaga	3840
agaugaauuc aaccucuucc agaaaauca uuuagaaagg aggagggagg aggcuaaacu	3900
uggaccuac aggaagucua ggcuuguaga agaggcgga uuaccugacu ggcuuguaaa	3960
gaauagcgau gagauugaga aguggacuua ugaagaaacc gagguccaaa uggaagagg	4020
uaauaggcag aggaaggaag uagauuauac agauaguug acugaaaaag aaugguuaaa	4080
ggccauugau gacaauugag augauuuuga ugacgaugaa gaggaagagg uaaaaacaaa	4140
gaaaagaggc aagagaagaa gaaggggaga ggaugaugaa gaagaugcaa guacuucaaa	4200
gagaaggaaa uauucuccau cugaaaacaa acugaggagg cguaugcgua accucaugaa	4260
cauuguuguu aaguauacug acagugacuc gagaguacuc agugaaccuu ucaugaaacu	4320
ucccucucgc cauaaguacc cagacuacua ugaguugauc aagaaaccua uagacaucaa	4380
gaggauauug gccaaaguag aagaguguaa auaugcugac auggaugaa uagaaaagga	4440
uuuuauugcaa cuuuuguaaa augcucagac auacaauag gaggccuau ugaucuauga	4500
agaauugaua guauuagaaa guguuuucuc uaaugcucgu caaaaaguag agcaggauaa	4560
ugauucagau gaugaugaaa guaaagguga ccaagaagau gcugcaucag acacucauc	4620
cgucaaaaug aaauugaaac uaaagccugg gaggaccga gggaguggag cugggguuaa	4680
aaggaggaga agaaaauaua ucucugaaga ugaagacgaa gaccuagcg aaguuccuu	4740
aauguaauc cucuucacug uccuuuguua uuauuaguu ucaucggugu ucgguaccug	4800
ucagucaagg gagaagcuua gcuuuuuagu ugacuauuga agaaauuagg acugaguucu	4860
guuuuuguuu uuuuuguuug uuuuuuuug gauaaaugua uuuaauagau aaaauguuc	4920
gcuuauauau auuuuuuuu cugguuuugu aaauggcc	4958

<210> SEQ ID NO 44

<211> LENGTH: 499

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 44

gaugaugaag aaugcaag uacuucacaa agagggaau auucuccauc ugaaaacaaa	60
cugaggaggc guaugcguaa ccucaugaac auuguuguua agauuacuga cagugacucg	120

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agaguacuca gugaaccauu caugaaacuu cccucucgcc auaaguaccc agacuacuu	180
gaguugauca agaaaccuau agacaucaag aggauauugg ccaaaguaga agaguguaaa	240
uauugugaca uggaugaaau agaaaaggau uuuaugcaac uuuguaaaaa ugcucagaca	300
uacaaugagg aggccucauu gaucuaugaa gauucgauag uauuagaaag uguuuucucu	360
aaugcucguc aaaaaguaga gcaggauauu gauucagaug augaugaaag uaaaggugac	420
caagaaugug cugcaucaga cacuucaucc gucaaaauga aaugaaacu aaagccuggg	480
aggacccgag ggaguggag	499

<210> SEQ ID NO 45

<211> LENGTH: 6346

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 45

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uguuuuuugc guugauaucg gugauuuuag ugaauauuag ugaaguguug auguuuuuu	120
ucuaauggcg ucugaagaag aaguugacga guguuuacca guugacgaug aaguugacac	180
uaguguuguu caacaagaag gcacugaaga aaauccaccu gacagugaug aaagaaguag	240
gauagaggaa gaagaugacg aguauacccc ugaggauugc aggaaaaaa agaaagguaa	300
aaagagaaaa gccaaagggg aaagcaaaaa agaaaagaaa cguaaaaaaa ggaagaagaa	360
ugauagugcu gaagaaagug agggaggcgg ggaagaagaa ggcgauuccg auuauaggag	420
aaaaucuaag aagucuaaag gaacuucaca accaaaacca gugcagcaag auucuuucug	480
agguguaccu ucaguagaag aaguuuugcag cuuuuuugga cuuacagaug uacagauuga	540
cuauaccgaa gaugauuacc aaaaucugac uacguauaaa cuuuuucaac aacauguucg	600
uccuauucuu gccaaagaca accagaaggu ucccuaucgga aaaaugauga ugcucguggc	660
ugcaaaaugg agagauuuuu gcaauuccaa uccaacgcu caacaggaa cagauccaga	720
agcuucagaa gaacaggaa auucuaaacc uaccaggaca cgaccuucac gaguuucaac	780
uacacaaaau gaugaugaag aagacgacga ugcugacgaa cgaggaggga aaaagagaag	840
uggacgaagu aaaaagucan caggaaagaa guccgcuccu ccggccacaa ccaagguccc	900
uaccucaaag aucaagauag gaaaaagaaa acagaaaucc gaugaagaag augaagguuc	960
aguuggugcc guuucugaaa gggacucaga ugcugaauuc gagcaauugc ucgcagaagc	1020
ugaagaaguu aaauaaccug aagguguugu agaagaagaa gaaggugcag agguggcucc	1080
uguaccuaag aaaaaggcca aaacgaaaau ugguauuaaa aagaaaagga aaaagacacg	1140
gacuacuaac aaguuuccag acagugaagc ugguuaugaa acagaucalc aggacuauug	1200
ugaaguugu caacaaggag gugaaauau auuauugau acgugccuc gagcuauca	1260
uuugugucgu uuggaucucc aaauuggaaga uacgccagaa ggcaauuggu caugcccuca	1320
uuugagaaggu gaagguguac aggaaaaaga agauauguc caucaagaau uuugcagagu	1380
uuguaaagau gguggagaac uuuuauugcug ugaauucugc cuucugcau accacacauu	1440
cuguuugaac ccuccauuga cagauauucc agauggugac uggaagugcc cacguuguuc	1500
ggcgaaagccu uugagaggua aagugucaaa gauucuuacu uggagguggu uggaauucucc	1560
caguagaaaa gaugaagaag acaauacuaa aaacgaaac aggcagaggc aaagagaaua	1620
uuucgucaag ugggcagaua ugucuuauug gcacugagu uggugugucug aacuuacgau	1680
ggauguuuuu cauacuaaaa ugaucaggag uuauauucgu aaauaugaua uggacgaacc	1740

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ucccaaacua	gaagaaccu	uggaugaagc	agacaauga	augaagagga	uacgagaggc	1800
aaauaucau	gagcaagaau	ugaagagaa	auauuacaag	uaugguauca	aaccagagug	1860
gcuuauugug	cagaggguaa	uuaaccaucg	cacuaaaagg	gauggaagca	aucuguaccu	1920
cgucaaaugg	agggaccucc	cuaaugacca	ggcgacuugg	gaggaagaag	ucaccgauau	1980
cccuggcug	aagaaagcua	uugaauuua	caaugagaug	agggcuugcu	guuuagguga	2040
aucuaaaaa	cuaaaaaaag	guaaagguaa	aagaucaaa	agagaucaa	augaugagga	2100
aggaagcaga	agugcaggaa	ugaugggcgu	cgguggacca	gcuacugguc	aaacuuccc	2160
gccuccugaa	aagccuguca	cagauuugaa	aaagaaauac	gauaaacagc	cggacuaucu	2220
cgacgucucc	gguaugugcc	uucauccua	ccaauuagaa	gguuuuuuu	gguugaggua	2280
uuccuggggg	caaggaacag	acacuaucuc	ugccgaugag	augggucucg	gaaaaaccu	2340
ucagacaaau	acuuuccucu	auucucuua	caaagagggg	cauuguaaag	gccccuuccu	2400
ugugagugua	cccuuauca	cauuuaucaa	uugggaaaga	gaguucgaaa	cuugggcgcc	2460
agacuucuc	guugucacau	augucggaga	caaagauucu	cgugcuguaa	uacgugaaaa	2520
ugaaauuua	uucgaugaua	augcuguuag	aggaggaaga	gguguuucua	aaguucgcuc	2580
uucugcaaua	aaguuucaug	uacugcuaac	aucuuaukaa	cuuauucua	ucgaugucac	2640
uugccuugga	ucgaucgagu	ggcgagugcu	uguaguagau	gaagcacaca	ggcugaaaag	2700
uaaucagagc	aaguucuuua	ggcuucucg	uucuuaccac	auugcuuua	aacuucugcu	2760
gacaggaacu	ccguugcaaa	acaauuaga	agaauuguuu	cauuuacua	auuuccuuac	2820
gccggaaaaa	uucacagacc	uugcgacuu	ucaaaccgaa	uucgcugaua	uuucaaaga	2880
agaacaaguc	aaaagacuuc	augaguuauc	cgggcccga	auguugagga	gauuaaaagc	2940
ugauguacuc	aagaauaugc	cuacaaauc	ugaguucuu	guuagaguug	aacucucccc	3000
gaugcagaag	aaguacuaca	aaauauuuc	cacaaggaau	uucgaagcuu	uaauccaaa	3060
aggaggcggg	caacaaguau	cucuuuugaa	cauuauaug	gaucuuaaaa	aaugcuguaa	3120
ucauccauac	cuguuuccug	cugcuucua	ggaagcuccu	uuaggacca	gcggauuuu	3180
cgaucuucaa	ggguuuauca	aagcaucug	aaaauugaua	cuucugucga	aaugcugag	3240
acggcucaaa	gaagaggguc	acagaguacu	gauuuucuc	caaauagcaa	aaauuguga	3300
cuuauuagaa	gacuaccucg	aggguagaag	uuauaaauu	gaacguauug	acgguacgau	3360
caccgguagc	uuuagacaag	aagcuauca	ucgguuuac	gccccuggag	cucaacuuu	3420
uguuuuucuu	uuguccacuc	gugcgaggag	ucuuuguuu	aaucucgcu	cugcagauac	3480
aguauuuuu	uauagacucg	acuggaauc	ucauacgau	auucaggccu	uuucgagagc	3540
acacaggaua	gggcaagcaa	acaagguuu	gaauuauca	uuugugacac	gagcgucugu	3600
ugaagaaaga	guaacgcaag	uggcuaagag	aaaaauaug	uuacccauc	uugucguacg	3660
accagguaug	gguggcaagc	aagcauuuu	cacuaagcaa	gaacuugaug	auuuuuuag	3720
guuuggaaca	gaagaacuuu	ucaaagaaga	gcaggguaaa	gaagaugaag	ccauucauu	3780
ugacgauaaa	gcuguugaag	aauuacuuga	ccggucgaag	augguuuug	aacagaaaga	3840
aaacugguc	aaugaauauc	uuuucuuuu	caaaguggca	aguuauuuu	cuaaagaaga	3900
agacgaagau	gaggaaauag	gaacagaggu	aaauaaacag	gaagcagaaa	auacagacc	3960
agcuuuuug	gucaaacugu	ugaggacca	uuauagcaa	caacaagagg	auuuuucucg	4020
aacucucggg	aaaggaaaaa	ggaauucgaa	acaggugaau	uacaucgacg	guggagugau	4080

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ggacucaaga	gagaacgccg	auucgacgug	gcaagacaac	cucucugacu	auaaucaga	4140
cuucucugcu	ccuucugaug	augacaagga	agacgaugac	uuugaugaga	aaaaugauga	4200
uggaacgaga	aagaagcgua	ggccagaaag	gagggaggac	aaagauaggc	cucuaccucc	4260
ucuucuuucc	cgagucggug	gaaacauuga	gguccugggg	uucaacgcc	gacagcguaa	4320
agcauucuuu	aaugcuauua	ugagguauug	aaugccaccu	caagaugcau	ucaacucgca	4380
guggcuuguu	cgagaccuga	gggguaaauc	ugagaagcau	uucagagcau	acguaucccu	4440
cuuuauagg	cauuugugug	agccugggcg	ggacaaugcc	gaaacauucg	cggauggugu	4500
uccaaggga	ggcuuaguc	ggcagcaugu	ucucacaagg	auagguguga	ugucacucau	4560
uaggaaaaag	guucaagaau	uugagcaauu	uaauggauau	uacucgaugc	cugaaauguu	4620
gaagaaacca	cuuguugaug	ccggaauugc	uaaaacaagu	gcuagcagua	uaggugaagg	4680
ugcuaguagu	uccgguacac	cugcaacauc	agcugcucca	aguccagcuc	cuacucuuuu	4740
ggauaagaca	caaaauaag	auuugaguga	aaaagaagau	ccgucaaaga	cugaagauaa	4800
aaccaccgau	gauuccaaac	ccucagaaga	ggcuaaagcu	gcagaugaug	caaaauagcc	4860
ucaggcguaa	ggagaaaagg	cagaaggau	uucuaaugca	aaccaaacuu	cugaagcuga	4920
aggaagcgau	gagaaaaaac	ccaaagaaga	accgauggau	guagauggug	aaggagaggc	4980
uaaagauagu	gauaagacag	aaaaacaaga	agguacugac	gaaaaaug	uagcccuaaa	5040
agaggaagaa	aaggauaag	aggucaacaa	agagaaggga	gaggaaacag	aggaaaagaa	5100
gguuaucgau	uuugaagaag	acaaaucuaa	aaggaaaauu	auguucuaa	ucgcugaugg	5160
aggauuuacu	gagcuccaua	ccuuauggca	aaugaagag	aaagcugcag	uaccugguag	5220
ggagucagag	aucuggcaua	ggaggcauga	cuauuggcug	uuggguggaa	ucguuaccca	5280
uggcuauuggu	cgguggcaag	auauucaaaa	ugauauuaga	uuugcuauua	ucaacgaacc	5340
auuuuagaug	gauguuggaa	aaggaaaauu	cuuagaaauu	aaaaauaa	uucuugccag	5400
gagguuuuag	cuucuuagc	aagcucuggu	gauugaagaa	caguuaagac	gugcagcuua	5460
uuuaaauucg	acgcaagauc	caaaucaccc	agcaauguca	cugaauagca	gauuugcaga	5520
gguuagaugu	cuagccgaau	cucaccaaca	ccucucgaag	gaaagucuu	cuggcaacaa	5580
accugcaaaa	gcaguguuac	auaauguauu	gaaccaauua	gaggagcuuc	ugcggauuu	5640
gaaaucugac	guauucgac	uaccagccac	ucuagccaga	auuccaccug	uagccagag	5700
gcuaacaug	ucugaacggu	caauacuuuc	uagguuggcu	gcaacuacuu	cuccugcgac	5760
gcccaccacg	ucccaucaa	cugguauug	aagcagucag	uucccugcug	gauuucuuuc	5820
agggcaguug	acuggaagcu	uuccgaaugc	caguuuuacc	aacuucaggc	cccaguuuuc	5880
aguuccuggg	caaacugcag	cccaggguuu	ucccgguauu	ugauaauga	aagcuggacg	5940
guaaauugcu	gcgagugaau	ucuccaugag	uaaaauaag	guuuuuuuu	uuuuuuaaga	6000
aagaaaaaaa	agaagcguuu	uguuuaguuu	uguugauagu	ucucuuuauu	ucuuucaaau	6060
uuguuuuagc	ggaaaaaaa	auguucuuu	uaaguaacuu	auaaaauugga	caugcuauuu	6120
aaauuuccua	uuaguuuuu	uuguuuuuu	uaaguuuuuc	gguauuguaa	gaaugucuau	6180
auguguaaga	gguguuacaa	gauugccuaa	auaccuugua	uuuuuuuuu	uuacuauuga	6240
auaaaaaaaa	aaaauuuu	acuucgaucu	uagguuaagg	guaaaaaaa	aaaauguuac	6300
uggaaaaaaa	aaagaaaaa	auaaaaaaga	uagccuuucc	ccuuac		6346

<210> SEQ ID NO 46

<211> LENGTH: 3391

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<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 46

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agagggggua ggcgcacagc uuuccucaca ucgaacaaua ucuuagugaa ugaauggcuu      60
uauuggcccg uucaaaauc uguuaaaugu ugguuugaua uauuuuuaua cuaacguuau      120
uuaacgcagc ucacaccaau aaaaauugcg aagccaaaug aaguuauguu ggauacaaca      180
gauacuguug aaaaauucua ugaauucucg ggagacacag agucguccaa ggguaaaaau      240
gaagauuuug aaacaaaaau ugaacugac cguucuaagaa gauuugagu ucuuguagaag      300
cagacagaaa uuuuuucaca uuuuauagaa aaucacagaa agucgaacag ccugcaaaag      360
ccuaaagucg gccgucuaag aaaggaaacu auaaaaugg caccagccgg uggugauggu      420
ucugccgacc aucggcaucg uagaccgag caggaagaag augaagaacu gcuugcugaa      480
aguaauacuu cuucaaaauc cuuagcaagg uuugacgcuu cuccuuuuua uauuaaaagc      540
ggagaguuga gggauuacca gauacuggu ugaauugga ugauauccu cuacgaacac      600
gguaauaaug guauacuugc ugaugagaug gguuaggua aaacucucca aacuaauucu      660
cuccuugguu acaugaagca uuauagaaau auaccagggc cacauauggu caucguacca      720
aaaucaacau uagcuauuug gaugaugaa uuuaaaaagu ggugcccaac ccugcgugcu      780
gucguuuuaa ucgggagaua ggaaacgagg aaugcguuca ucagagacac ucuuugccg      840
ggugaauagg augucugcg uacaucuau gaaaugaua uacgagaaaa gagcguuuuc      900
aagaaguuca acuggaggua uauggucauu gacgaagccc acaggaucaa gaaugaaaaa      960
uccaaacucu ccgagauugu gagagaguuc aaaacgacga aucgauuacu ccugaccggu      1020
acuccuuuac aaaaauaccu ccacgaauug uggucucuuc uuaacuuccu cuuaccagau      1080
guuuucaauu caucagauga uuugauuca ugguuuaaua ccaauaccu ccuugcgau      1140
aaucucuuug ucgagagauu acaugcugua cugagaccu uccuccuag aagauugaaa      1200
ucugagguag agaaaaacu caaacgaag aaagaaguca aaauacuag uggauugagu      1260
aaaaugcaga gagaauugga uacuaaagu cuauugaag auauagacau uguaaacggu      1320
gcuggccgag ucgaaaaaua ggcgccucca aacauccuca ugcaguugag gaagugcagu      1380
aaucacccuu aucucuucga cggagcugaa ccagguccac cuuacucaac ugaugagcau      1440
cugguauaua acaguggaaa aaugguaua uuagacaagc ucuuccuaa auugcaagaa      1500
caaggauac gaguucuggu uuucagccaa augacaagga ugaugauau ucucgaagau      1560
uacuguuauu ggagaggaua uaaauacugu cgucuugaug guaaucacc ucaugaggau      1620
aggcagagac agauuaaaga guucaacgaa gaagacagua agaaauucau uuucauguug      1680
ucgacucgug cgggugguuu ggguaucuu uuagccaccg cagauguagu cauuuuguac      1740
gauucggauu ggaacccuca aauggaucuc caggcuagg aucgugcuca ucguauuggu      1800
caaaagaaac aagucaaagu guucaggaug auaacugaaa acacaguuga agagaaaaau      1860
guugagagag cugaaauaaa acuccgccuc gaaaguugg ucauccaaca aggcaggcug      1920
guagacaaua aaacggcacu caacaagau gaauguuga auaugauccg ucacggugcc      1980
aaucauguau uugccaguuu agauucugaa aucaccgaug aagacauuga cacuaauuug      2040
gaaaaggcg aagcaaggac ggaagaaug auaaaaaaac uugaacaacu cgguauucu      2100
auuuugaag acuucaugau ggaaccccg acugagucag uuuccaaau cgaaggagag      2160
gauuacaggg aaaagcagaa aguuuagga auaggaguu ggauagaacc uccaaaaaga      2220

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gaacguaaag	cuaauuacgc	ugucgaugcc	uaauuuaggg	aagcauugag	aguaucagaa	2280
ccuaaagcuc	ccaaggcacc	gaggccuccu	aaacagccua	uaguucaaga	uuuccaauc	2340
uuuccuccuc	gucucuuga	gcuauggac	caggagauuc	auuacuucag	gaaaacugug	2400
ggcuacaaag	uuccuaaaaa	uccugaauua	gguucugaug	caucacgugu	ccaaaaggaa	2460
gaacaaagaa	agauagauga	ggcagaaccu	uuaucaaga	aagaacucgc	ugaaaaggaa	2520
aaacuucuaa	cgcaggguuu	uaccaauugg	acuaaaagag	auuucaccca	guuuuuuaa	2580
gcuaaugaaa	aaauuggucg	ugaugauuuu	gacaauuuu	caaaagaagu	agaaggaaaa	2640
acuccagaag	aaguaagagc	uuauucagaa	guguucuggg	aacgauguaa	cgaauugcag	2700
gacauagauc	guaucauggg	gcagaucgac	aggggagagg	cuaaaauuca	aaggagagca	2760
aguauuuaga	aagcucucga	uacaaagaug	agccgguaa	gagccccauu	ucaucaacuu	2820
cgaucuccu	acgguacgaa	uaaggguuag	aacuauaccg	aggaagaaga	uagauuccuu	2880
gucuguaugu	ugcauaagcu	ugguuuugac	aaggaaaaug	uguacgaaga	acuuagagcg	2940
auggucaggu	gugcgccuca	guucagauuc	gacugguuca	ucaaaucgag	aacagccaug	3000
gaauugcaga	ggcguuguaa	uacucuaauu	acucucaucg	aaagagaaaa	ucagggaacuu	3060
gaggagaggg	aaagagccga	gaagaggaaa	ggaagaggaa	gugggagugg	uccugguucc	3120
gguaaaagga	aaggagacgg	uuccauuua	ucucccccuc	cugucccugg	ccaaggggau	3180
aagaacagcc	ccgccagaaa	aaagaaaaaa	auguaguuuu	accucccau	gaaaggaacu	3240
cauuuuuaga	uauuuuuuc	uagauuuua	uuuugugaaa	acugugaugu	auuuuauuc	3300
cguuccgaaa	agcucuacug	uuuugacagu	uuuuuuuuu	aguggggugg	ggaggaaaua	3360
uagccccuc	accccccau	aaucuaaaa	u			3391

<210> SEQ ID NO 47

<211> LENGTH: 1316

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 47

aaugaaaaa	aaacuugaac	aacuuggugu	ugauucauca	uuaaaagauu	ucaugaugga	60
ggcucccacu	gagucugucu	aucaguuga	aggcgaagau	uauagagaaa	agcaaaaagu	120
uuuuggaauu	ggaaaaugga	uugaaccacc	aaaacgagaa	cguaaagcaa	auuaugcagu	180
agaugccuau	uuuagagaag	cacugagagu	uucagaaccu	aaagcuccaa	aggcccuag	240
gccaccaaag	caaccuauag	uucaagauuu	ccauuuuuu	ccaccucguc	uguuugagcu	300
guuagaucaa	gaaauuauuc	auuuucgaaa	aacuguuugc	uacaagguuc	cuaaaaaucc	360
ggaguuaagg	ucagaugcuu	cucguauaca	aagggaagag	caaagaaaa	uugaugaagc	420
ugagccguug	acugaggaag	agcuagcuga	gaaagaaaac	uuauugacc	aggguuuuac	480
uaauuggacu	aaaagagaau	uuuaccaguu	cauuaaaagc	aaugaaaaau	auggacguga	540
ugauauugau	aaauucucua	aagauguuga	agggaagacu	ccagaagaag	uacgagcaua	600
cucugaagua	uuuugggaaa	ggugcaauga	acuacaggcc	auagaucgua	ucauggggca	660
gauugauaga	ggugaagcga	aaauucaaa	aagagccagu	auuaaaaaag	cuuuagauac	720
aaagaugagu	cgaauuagag	caccguuua	ucaacuacga	auugcuuug	guacgaacaa	780
ggggaaaaau	uacacagaag	aagaagacag	auuccuugug	ugcaugcuac	auaagcuugg	840
cuuugauaaa	gaaaaugugu	augaggaacu	uagggcgau	gugaggugug	cuccucaguu	900
uagguuugau	ugguucauca	agucucgaac	agcuuuggaa	uugcaaagac	guuguauuac	960

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ucuaaucacg uuaauugaaa gggaaaacca agaauuagaa gaaagggaaa aaguagaaaa	1020
aaggaaaagu cgaggcagua augggcgugg ucccaguucu gguaaacgua agggagaugg	1080
aucuaauuca ucuccaccug ucucuguaca gagugauaaa agcagcccug cucggaaaaa	1140
gaaaaaguuu aucucuguug aguaaaauua ucuuaaaacu gggaguagau acccaauucu	1200
cauuauccgg ugaucaagga aucaaucuca uauaggagcc uaaaacuua uuaguuuua	1260
auugaauuu uaaauuacau cucuaguuc caaaauuugu uucuuuuaca ucugua	1316

<210> SEQ ID NO 48

<211> LENGTH: 1827

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 48

gauaaaaug auaagaaaa uuuuuuuuu auuuguuuca uaaaaaaaau aucuuauagg	60
uuuuuugauu auaauuggu ucaaucauaa aaucagagau acuaagauu guuuuaucau	120
aacaaaccca aucucuagua ucgucauccu gcuguucugg uucacucuga guuuuuuuu	180
cuucaucaa agcaaaacu gcaacuuaa aagcagaaag uaaaucauca ccaacagugg	240
cugguccuuc aucucugguu ucagcucuuc ucaaaauuuc gucaauguca caaguugguu	300
cuucaucacc aucucuuca ucuuuuuuu auucucagc cccaaauuuu aaaaagcag	360
uaaguucuu uuuguuaaaa ggcgcacugg augaagauu uuuuuuaucc aggacaguuc	420
uaccguagu auccauucu uguauaacua aaugaucua gaccuuuuu uguuuggcc	480
gcucgacaa auuuuccuca acagaacuu uaguaacaag ucuguauaug uucaccugau	540
uuuucugacc gauucauga gcucuagcu gugcuugca aucauuuuu ggauuccaa	600
cagagucaaa uauaaugaca guaucagcug ugcuaaaau aaugcccaa ccaccagc	660
gaguugauaa uaagaaacag aaucuggug auuuuucagc auugaauga ucgagggcuu	720
gcuuucucua uucaccuuu auugaaccgu cuaaacguug gaaagggaaa ugucucauu	780
gaagauacuc agccagaua uccaacauuc guaccuuug agaaaauua aguacucua	840
gccaguuuc uuuaggcga acaagcaacu uguccaacag aaguauuuuc ccugagccuu	900
uuacaaauug cuguuagug ucucaguuu uugcuucau uucuaauggu uuuaauagau	960
gugcaugau acagcauuuu uuuuuucaa uaacaauuu uauaaugua cuaggagaac	1020
cuuugacucc uuucgaaga gcagaauuu uuugggaca aaucaccug uauucugcu	1080
ucuguacaga ugucuuuca acacguaua uuuguuccac uuugcuggu aaagauucu	1140
caacauccu cuuacucgu cguagaauu augguuccag cugucuguc aacuuaguau	1200
agccuuuuu agcagaguug ucauguucu uuucuuuuu uucccaguu uaaaucugu	1260
ugggcauuu aaagugaagc aacgcccua gcucuuaag acuuuuuugc aaaggagugc	1320
cuguuauaag aagccuagg uuuguauca acucuucua uguuuuguu aauaauaau	1380
cauauuuuu caucugugu gcuuucaua ccauaaggau agccagcuu auacuacca	1440
aaaaugcuu gucuuuuaga acauuuucu auguaguaag aauggcuuu aauuuuacc	1500
uuuucgaacc ugaauagc acuuucauu uacguauaac aucacgggag uuuaauacac	1560
caauuaagu uacaacauu auuucuggag ccuaaaua aaacuccuc ugccaugaag	1620
ucaucguaga uaaagggaca acauuuuuu augguccau caacugguga guaugaaau	1680
aaauuacaa acugcagaua gucugaauug uuuuaccaag acccauuuca ucagccaaa	1740

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uaauagaaau uucuuuacac cacgaugaa ccaaccaauu caaaccacug auuugauaaau	1800
cucucaaaac caauaccugg ucaccac	1827

<210> SEQ ID NO 49
 <211> LENGTH: 496
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 49

gacuaccucg agggugaagg uuauaaaauu gaacguauug acgguacgau caccgguagc	60
uuagacaag aagcuauca ucgguuuuaac gcccugagg cucaacaauu uguuuuuuuu	120
uuguccacuc gugcgaggagg ucuugguuuu aaucucgcuu cugcagauac aguuuuuuuu	180
uaugacucug acuggaauc ucauaacgau auucaggccu uuucgagagc acacaggaua	240
gggcaagcaa acaagguuau gauuuuauca uuugugacac gagcgucugu ugaagaaaga	300
guaacgcaag uggcuagag aaaaaugaug uuaacccauc uugucguacg accagguaug	360
gguggcaagc aagcaauuuu cacuaagcaa gaacuugaug auauuuuaag guuuggaaca	420
gaagaacuuu ucaagaaga gcaggguaaa gaagaugaag ccauucuuu ugacgaaaa	480
gcuguugaag aauuac	496

<210> SEQ ID NO 50
 <211> LENGTH: 481
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 50

caaaaaauga aacugaccgu ucuagaagau ugaguuucu guugaagcag acagaaaauu	60
uuucacauuu uaugacaaau caaggaaagu cgaacagccc ugcaaagccu aaagucggcc	120
guccuagaaa ggaacuaau aaauuggcac cagccggugg ugaugguucu gccgaccuac	180
ggcaucguau gaccgagcag gaagaagaug aagaacugcu ugcugaaagu aaucuuucu	240
caaaaucuu agcaagguuu gacgcuuuc cuuuuuuuu uaaaagcgga gaguugaggg	300
auuaccagau acgugguuug aauggauga uauccucua cgaacacggu auaauggua	360
uacugcuga ugagauuggu uuagguaaaa cucuccaac uauuucucuc cuugguuaca	420
ugaagcauuu uagaaauua ccagggccac auauggucau cguacaaaa ucaacauuag	480
c	481

<210> SEQ ID NO 51
 <211> LENGTH: 490
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 51

guucaagauu uccaaaauuu cccaccucgu cuguuugagc uguuagauca agaaauuac	60
uuuuucgaa aaacuguuug cuacaagguu ccuaaaaauc cggaguuaagg aucagaugcu	120
ucucguauac aaagggaaga gcaagaaaa auugaugaag cugagccguu gacugaggaa	180
gagcuagcug agaaagaaaa cuuauugacc caggguuuua cuauuugac uaaaagagau	240
uuuaaccagu ucauaaaagc uaaugaaaaa uauggacgug augauuuga uauuauuca	300
aaagauguug aagggaagac uccagaagaa guacgagcau acucugaagu auuuugggaa	360
aggugcaaug aacuaacaggc cauagaucgu aucauggggc agauugauag aggugaagcg	420
aaaauucaaa gaagagccag uauuaaaaaa gcuuuagaua caaagaugag ucgauuuga	480

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gcaccguuuc 490

<210> SEQ ID NO 52
 <211> LENGTH: 496
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 52

cagcuggaac cauaauuucu acgacgaguu aagaaggauug uugagaaauc uuuccagcu	60
aaaguggaac aaauuuuacg uguugaaaug acaucuguac agaagcagua uuacaggugg	120
auuuugucca aaaaaauuc ugcucuucga aaaggaguca aagguucucc uaguacauuu	180
auaaaauuug uuauugaaau aaaaaaagc uguaaucaug cacaucuaau aaaaccauuu	240
gaaaauagaag caaaaacuga agacuacuua cagcaauugu uaaaaggcuc agggaaaauu	300
cuucuguugg acaaguugcu uguucgccuu aaagaaacug ggcauagagu acuuauuuu	360
ucuaaaugg uacgaauugu ggauuuacug gcugaguauc uucaauagag acuuuuccu	420
uuccaacguu uagacggguu aauuaaaggu gaauugagaa agcaagccu cgaucauuu	480
aaugcugaaa auucac	496

<210> SEQ ID NO 53
 <211> LENGTH: 4493
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 53

gcuaauacag aaacaaucaa aagaucuuu cuuacucguu caaacagcag gaugaagaaa	60
acuaggaaga auucucgacc uuuaauugug gcuggugguu uugcagcggc agcugagaag	120
augccugagu aggccugauu gagagugcag guacggauug cuuucuccc gguccucua	180
guccucuggg gccacgugag ccaauaugac cagggccgac gggccuacac cguggccucc	240
cuggaccucg acgaauugcu cgguugcuua aagcaggcau gaaaggccuc gaaauuuuug	300
guggugucug gcugauaggu uccuccuggu cgguguuguu aucauaggag ucuuggcucg	360
auuucauac guccgugccc uucggacucu cagcugguga uuucacaccu ugguaaacuu	420
caacgacccc auuucaguc uucauauuu ucaccguguc cuuguaauuu ccuagagcau	480
uuuuucuuuu ucuuucucu ucccuugaga uaucuugccu ugaauucua aguugucucc	540
ucucugcuug cuucugccga uauuucuguu cuauuucugc aucaucaagc aaauagagaa	600
caaccucuuu cgguuuaaga gugucugguu ugaaauuacc cccacugaug acaaccuuu	660
ggauuucacu uuuuucuuu gcucucugca agauccuuc ucgauagguc cccuuacaaa	720
ugagccgaa gacugugacu uguuucguuu guccaagacg augggcacgg uccaucgcu	780
guuggucaac aguaggguuc caauacugc cauagaauu cacaguauc gcugcgguua	840
ggugauuucc aagaccucca gcucgcuac ucaacaggaa uacaaauaug uccucucuug	900
uuugaaguc agcaaccau ucccuucugu cugauaucuu ugaagaacca ucuuaccuca	960
uauaugugug cuuucuguc cacauauuu ccucuaacaa gucgaucac cuugucauc	1020
gggaguaauu uagagcucga ugcccuuguu ccuucagccu ggugagaagc ccauccaaga	1080
cguacagcuu uccagcaua guuaccagug ucuguuuguc agguaugacu auacucgacc	1140
agccagugau uggucgaaga cugaguaggc caagaggagg ugggcacugg aaccagguu	1200
cuuguuuuac ccuucccaa agaaccugcc acagucuccc guccuccccc cagaggugag	1260

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aagagaucca	gcuccaccuu	cguucgagc	uguacagccg	ucugcuuuuc	uuuauccca	1320
ccuugacagu	aggagugaag	aggaauggag	guaaaagac	uggcugacau	gagagaauuc	1380
gaggcuuccu	cacaaugu	ggcaaugcug	guaauaagu	agcacuauuc	ucuccaccgc	1440
acucucccuu	agccuucuu	gaccugacua	ugcgauuuuc	uauccguuuc	ggcauacuuu	1500
cuaucccac	gguggaagg	gaaaacacuu	gguuggaaua	aguggugaac	acaagauugu	1560
uuggguagcu	ccugucuuca	augauuccga	cgaguuuuccu	ucucacuccu	ugccuccuug	1620
cuauauuuu	gugccaccug	aaaauaaucc	caaaaaacau	uagcuuauac	aguucuccua	1680
cagauaaccu	caacagucga	gugaaagaaa	augaacuauuc	ccucucggau	gagucaucac	1740
ugaauaaaga	ucuaugagaa	ugaaaaggau	uaaaggcgga	uaaguauuc	augagaagau	1800
gaaguuuau	cccagggaac	acagcaucug	ugaugagagc	aggcacaau	uaucuuuccg	1860
uagccaugga	aaaaggggau	cgagguuucc	uucgcucgaa	uaguucaggg	ugauuacaga	1920
ccuuacgaaa	uugcaucacg	agguucauca	aaauugaagu	gauacucuga	gcugauuggu	1980
aagaagaucc	agaagagugu	agcaauucuu	caauucgaau	cuucuuuuuc	acagcugaau	2040
auaacaucuu	cugccucguc	gucagaggac	aguacaccau	gauuucuaau	uuauucgaca	2100
guucauucuc	cacaucuguu	uuuacucucc	gcaacaugaa	ugguuuuagg	aucauangua	2160
aacgggacaa	augcuuuuca	ucaauacugg	uuuuauugcuc	ugcaugacuu	ucuauaucuu	2220
uugaaaacca	uucuuugaac	ucaucgugug	aaucacaacu	ugagggcauu	augaaaugaa	2280
gaagagccca	aaguuacagc	auugaguuuu	gaauaggugu	uccacucaga	aguaauucugu	2340
ugcggaauu	gaauccaaga	agcaauuucc	aacgcaugcu	uguagugcuu	uugauagccu	2400
gagcuucguc	uagaauuaaa	uacugccauu	uuauccuauu	gaaguauuuu	auaucaguua	2460
uuacaagcug	auagcuugug	aucacaacau	ggaaacuggc	aucuuuagua	uguuaaccuu	2520
uuugauccca	aaauugacgu	aaauuuuucc	uuuccugcug	auuucccaa	uaaggcaca	2580
cuuugaaauc	agguaaaaa	cgcugcauuu	cuugcugcca	auuauguauu	guagaagcgg	2640
gcgauuuuu	gagaaaugga	ccccaacag	aguauuuuuc	agcaauaugg	caaagaaagg	2700
cuaucgauug	gacugucuuu	cccaauccca	uuucaucugc	caagauucca	uuauuaccuu	2760
ggucauauaa	auucacaagc	caugucuuu	ccuuuauuug	auaucccuug	agaguaccac	2820
ggaauaucug	cggguugggu	uuauucucac	caacauucc	auccucuucc	auuuucggc	2880
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guacuuuuuc	uuucucugcc	ucgcaauucg	agucauccaa	agguaaggag	cuaggauuag	3000
cuuccucuu	aaguuggcuc	aggauucgaa	guugaucuuc	agucguuccg	ccaccgagcu	3060
uacgggacau	aaagugagca	uagaguucug	uuugaguauu	gaggaaauu	aauuuucuuu	3120
gcugcccuu	agccuccauc	aguucuaacu	ccaacuuccu	uuguuccucu	gcuuucuuu	3180
ccauucuuu	ucuuuuuucc	cuuuccaccc	ucucaaaucu	uuuccaguuu	acuugcauuu	3240
cccguugcaa	ucuuuuugcu	cuccaaaaua	ccucuuucau	auucuuuugc	gauugcauug	3300
cacguugcug	acagugccuc	auacaguua	uagcagcucg	ccugcaagcu	guuagaaauu	3360
cuuuauuggu	acuuauccua	uagcgugaa	ccuuuccaau	uucuuuuuuc	gccauugug	3420
cccauuuuu	acgccugcga	ugugccauga	uuucggcagc	uuuguuuug	gcugauuuu	3480
uucuaaggu	cauuuccuuu	uuagaccuug	acauuuuuug	uaggucuggu	ucuuucuuu	3540
uuuuucuuu	cuuuuuuuc	acacuaaaac	uuucaugug	uugaucuuu	uuuuucaacu	3600
uuucuuuuu	uugaaaaaca	aauuucuuu	ucuaacaaa	accagaacu	gaacucugau	3660

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gcucaggaua cuuauacgaaa uuggagagga guccugugcc auaguacaua uacucuuggu 3720
uuuuagaauu gugguagaau uuuuuuuuau auuuguuucu aaguacaugu ucacggagca 3780
ugucuuguua guccucuua guuuuuuuu caucaucaga ggaauucgau gugucaguca 3840
auagaacauc aacuaaccac ugccuauua agcuuacau acucaaguug uaaagccucu 3900
ucuugucagc uauccuauu uuuuuaguug cuguuauacc auuccauacc guuucuccgg 3960
uauaggcauc aacaccagcu aaauccgugu cagaauacau ggaggcuucu ccauccucac 4020
cuauaggguug uuuuaaaaag ucuucaacau auguuaggaa aggagcuauug uccaagcuuu 4080
uuucuagcuu uugauaauaa agagguuugg caauuucugu uuucacuacc auguuuguuu 4140
uaucaucacu cauacugcaa aaaucaauga caucaaaaga uaucucugca ucccaguua 4200
aaaaauuac uaaacugaaa uguaaaacuu auauagaauc auuuuuuuuu ugaguugaac 4260
aaacuauuac gcuugucaca uuuuuaguua accacaccca aaauaauauc uacuuuuuua 4320
cauaaacua aucagaauu cagucagucc auacuagacg auuguaaaaa ugugcuaggg 4380
gucaaaaaa aaaggaaagu gaaauuaggu uaguauauu ugaaagacgc aucuccuuuu 4440
cagagauuca gugaaauuu ucagccagcu gguuagccu gacagaauuc aag 4493

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<210> SEQ ID NO 54

<211> LENGTH: 6108

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 54

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uugauuguga aaaaauuua agcuuugaau ccuuauuuu aaauuuuac uauuuaccgg 180
caguguauca ugaagcuaaa cuuguacuua uagucaaugg aacaacauu gauggacgcu 240
uuguuuuacu uaaccgacca acuuucugc ugaccuuacu aaucuaaua cuugaggcag 300
cagaagcucu ucuaguacga auuacaaggu uaggguucga auaggguugu gggggaggau 360
uaggaggagg gggaggguua cuacgccguu uaccacucu aaauuuuua cuuugcccug 420
augaggguuu ccccccuuu ugaggacugu caucuaaagu ccacagaua auugaaacac 480
gaccaugaga ucuaguccu ggagugcccu ccucgauacu acuccacua ugacauuuu 540
uuccccccg cccacuagcu cuuguuuuu ugaccugauu ucgcgaauc acgccagagu 600
aagugagaac agcuuccgac ucagcuuccc cagaagguga uuuuuauu cuuugcagag 660
cagccagucu accagcucc cacucuuuuu uuuguuguu gauuucugcu ucagcugcag 720
ccaacugcuc uuuuagccaa gcagcaucau uuucuuucca gaauuuuau gcuaaccucu 780
caacagcaga aagcuguugc auuagauugu gaaguucua uucugccuug cucauuuucu 840
gcccaacucc uucaugggua ucaauaggua uuuuuuauc gaacucugcc aacucagcag 900
cagcuucugc uuuaagcaacc uggcugccg caacaucgga uucauccua gcuuuggcca 960
uugcacucuc gagcgacca auugcuacu ucucaucaga guucgcaacc aucugugugu 1020
cuucaggauu cugagcaguu uuaucacuau uaugaagaac uucugcauc cuucgggaag 1080
caucauucuc ggagguguca acauuuuaa gaucuuuga uuguuagcuc uuaaaguuaag 1140
cuguaguuu guuuccuccu ucaauaggua caucucccag cauucucuuu ugguucgcuu 1200
uuuuuaguu auuuuccua acaguuuuu cgcugaucaa ucuauuuuu uguuacauuc 1260

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ucguuuggcc	gauucugugg	caucgaucuu	gagcuugggc	auccauggua	ggauuccaau	1320
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uuguggauag	aaauaagcaa	aaauuucucu	ugucagcauu	aaagcgaucc	auuaagagcu	1440
gucucugauc	uacuuucguu	guuccaucaa	gacgaagaua	uaugugcccg	ugaaaguuaa	1500
gaaaugcuuc	caguacgucu	aacauucua	ucauuugugu	aaauuuuuu	auccgaugau	1560
ggucagcuuu	caaucuucga	agaagcuugu	cuaaugauug	aagcuuucca	cagucuaucu	1620
guauuagucu	ucuaucaggg	aacugcgua	ucauugcuga	ugauaugcua	ugaaguaauc	1680
gaaguuuagg	ucuuagccaa	guaucaacua	augauagccu	cuucucuucu	uggaacaucu	1740
uugaaggugg	cggauugggc	acauguaaac	ggacagguug	gcuggaaaca	gccgguacgu	1800
acaaaacaaa	ccuagagaau	auaucagaca	acuccgcaac	ucggucuuucg	auggaaugaa	1860
uagcugcugu	gagagcauga	gucugauucc	aaaauaaca	gggguuugaa	gauaaagcau	1920
ucuuacagug	aacguuucca	augcagucuu	uaguuucauc	aggagcauca	uccaauuuu	1980
aagaugauag	aaggucagaa	ccauaaaau	gaagggccug	acaccuuugu	ucauugauuc	2040
uaacaucag	uuccaacuuc	uccuuucgcc	guuuuuuucu	aagauuuucu	agguauuucu	2100
cucucaaauc	uucuuucgga	uuagauucuu	cuuuguuuuu	uuuuuuugag	uucaaaguac	2160
uuugucuugu	uacuuuuuuu	agagaaggua	augaauugac	guucgaucca	uuuccuugag	2220
cuggcucuga	cgauaugcuc	acaggugaau	uguuuacagu	agucaaaacug	uuugaacaa	2280
caguagcugc	agaagugaug	uuuaauggag	guaccucacu	gauaggccga	uuugaauuu	2340
uuugggucac	agaaguagg	uugacuaguu	uagcuacuu	gcuucccugu	uuggauacca	2400
caguuaucuu	uugcccagua	guagucuuu	caguagucag	accuccuuga	ggaguauua	2460
ccgguuuggg	agagaguacc	agcugccuac	cgguaggggg	auuaacuaau	ugugcauuu	2520
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uaacuuguc	aaugggccua	acagggcucag	aaaccugagg	ugguuugggc	aaauaccguu	2820
ucuuagacuu	aauuuuuacc	cuuccuuuag	ggcaaggugg	uagauagga	ggugcagaau	2880
caauugugcg	aaugaaguc	ggauuaacuu	uguauuuacg	agcucuguga	gcaacaaug	2940
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accagaacug	uuugauuuu	uucgcauuu	aagaagcuau	gcguuuuac	ugcauuucuu	4920
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auucugcuuu	gguaggacua	gcagauaug	gaaccgucac	aucaguauug	cuuccuggcc	5340
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cuggcuuuuc	aaagccaagu	ugaagauugu	ucauuucaa	uucuuuugu	uaucauuu	5940
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<210> SEQ ID NO 55
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 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
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 <220> FEATURE:
 <221> NAME/KEY: misc_feature
 <222> LOCATION: (216)..(216)
 <223> OTHER INFORMATION: n is a, c, g, or u
 <220> FEATURE:
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 <222> LOCATION: (222)..(222)
 <223> OTHER INFORMATION: n is a, c, g, or u

<400> SEQUENCE: 55

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cchuuygcha cmacbgmgga raarguygar yudaaygarg argaracvau yyukauiay 180

mgdcguyudc ayaarguyyu kcgwccuuy yuvyunmgdc gnyuvaaaaa rgargumgar 240

<210> SEQ ID NO 56
 <211> LENGTH: 27
 <212> TYPE: RNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2/Helicase degenerate dsRNA sequence
 <220> FEATURE:
 <221> NAME/KEY: misc_feature
 <222> LOCATION: (21)..(21)
 <223> OTHER INFORMATION: n is a, c, g, or u

<400> SEQUENCE: 56

mghgcygubu gyyuhauygg ngaycar 27

<210> SEQ ID NO 57
 <211> LENGTH: 60
 <212> TYPE: RNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2/Helicase degenerate dsRNA sequence

<400> SEQUENCE: 57

uayaarcuyc uvyusacmgg machccgyub caraacaayc umgargaryu ruuycauyur 60

<210> SEQ ID NO 58
 <211> LENGTH: 61
 <212> TYPE: RNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: SNF2/Helicase degenerate dsRNA sequence

<400> SEQUENCE: 58

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g 61

<210> SEQ ID NO 59
 <211> LENGTH: 23
 <212> TYPE: RNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Bromodomain degenerate dsRNA sequence

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<400> SEQUENCE: 59

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<210> SEQ ID NO 60

<211> LENGTH: 65

<212> TYPE: RNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: HAND-SLIDE degenerate dsRNA sequence

<400> SEQUENCE: 60

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cchmg 65

<210> SEQ ID NO 61

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<223> OTHER INFORMATION: Chromodomain degenerate dsRNA sequence

<220> FEATURE:

<221> NAME/KEY: misc_feature

<222> LOCATION: (33)..(33)

<223> OTHER INFORMATION: n is a, c, g, or u

<400> SEQUENCE: 61

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<210> SEQ ID NO 62

<211> LENGTH: 56

<212> TYPE: RNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

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<220> FEATURE:

<221> NAME/KEY: misc_feature

<222> LOCATION: (29)..(29)

<223> OTHER INFORMATION: n is a, c, g, or u

<400> SEQUENCE: 62

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<210> SEQ ID NO 63

<211> LENGTH: 4569

<212> TYPE: DNA

<213> ORGANISM: Euchistus heros

<400> SEQUENCE: 63

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ggttaccaag gagggatgcc acacatgaat ggaccaaagtg gtgttcctcc tggatatgcag 240

caggctactc aaacatttca gctcatcag caattgccac cccaccagca accaccaatg 300

cagactgctc ctggtggggc tgctagtggg ggaggacaag aaaatcttag cgctctccag 360

cgtgcaatag attctatgga agagaaaagg cttcaggaag atccacgtta ctgcgagctg 420

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gttgggggtg ggttcagcgg tgagggtggg gccctcctc ctgctaaca cagcttcagc 540

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<211> LENGTH: 6222

<212> TYPE: DNA

<213> ORGANISM: Euchistus heros

<400> SEQUENCE: 64

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gaggagaag atgacgagta tgacctgag gatgcgagga aaaaaagaa aggtaaaaag	180
agaaaagcca aaggggaaag caaaaaagaa aagaaacgta aaaaaaggaa gaagaatgat	240
agtgtgaag aaagtgaggg aggcggggaa gaagaaggcg attccgatta tggaagaaaa	300
tctaagaagt ctaagggaac ttcacaacca aaaccagtgc agcaagattc ttctggaggt	360
gtaccttcag tagaagaagt ttgcagcctt ttggactta cagatgtaca gattgactat	420
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<210> SEQ ID NO 65

<211> LENGTH: 3072

<212> TYPE: DNA

<213> ORGANISM: Euchistus heros

<400> SEQUENCE: 65

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actgaccgtt ctagaagatt tgagtttctg ttgaagcaga cagaaatttt ttacatttt	180
atgacaaatc aaggaaagtc gaacagccct gcaaagccta aagtcggccg tcctagaaag	240
gaaactaata aattggcacc agccggtggt gatggttctg ccgaccatcg gcacgtatg	300
accgagcagg aagaagatga agaactgctt gctgaaagta atacttcttc aaaatcctta	360
gcaaggtttg acgcttctcc tttttatatt aaaagcggag agttgaggga ttaccagata	420
cgtggtttga attggatgat atccctctac gaacacggta taaatggtat acttgctgat	480
gagatgggtt taggtaaaac tctccaaact atttctctcc ttggttacat gaagcattat	540
agaaatatac cagggccaca tatggtcac gtacccaaat caacattagc taattggatg	600
aatgaattta aaaagtgggt cccaaccctg cgtgctgtct gtttaatcgg agatcaggaa	660
acgaggaatg cgttcacag agacactctt atgccgggtg aatgggatgt ctgcgttaca	720
tcttatgaaa tgatcatacg agaaaagagc gttttcaaga agttcaactg gaggtatatg	780
gtcattgacg aagccacag gatcaagaat gaaaaatcca aactctccga gattgtgaga	840
gagttcaaaa cgacgaatcg attactctg accggtactc ctttacaaaa taacctccac	900
gaattgtggt ctcttcttaa ctctctctta ccagatgttt tcaattcatc agatgatttt	960
gattcatggt ttaataccaa taccttctt gccgataatt ctcttgatga gagattacat	1020
gctgtactga gaccttctc cctaagaaga ttgaaatctg aggtagagaa aaaactcaaa	1080
ccgaagaaag aagtcaaaat ctacgttga ttgagtaaaa tgcagagaga atggtatact	1140

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aaagttctaa tgaaagatat agacattgta aacggtgctg gccgagtcga aaaaatgcgc	1200
ctccaaaaca tctcatgca gttgaggaag tgcagtaatc acccttatct ctccgacgga	1260
gctgaaccag gtccacctta ctcaactgat gagcatctgg tatataacag tggaaaaatg	1320
gtaatattag acaagcttct tctaaattg caagaacaag gatcacgagt tctggttttc	1380
agccaaatga caaggatgat tgatattctc gaagattact gttattggag aggatataat	1440
tactgtcgtc ttgatggtaa tacacctcat gaggataggc agagacagat taatgagttc	1500
aacgaagaag acagtaagaa attcattttc atgttgctga ctctgctggg tggtttgggt	1560
atcaatttag ccaccgcaga tgtagtcatt ttgtacgatt cggattggaa ccctcaaatg	1620
gatctccagg ctatggatcg tgctcatcgt attggtcaaa agaaacaagt caaagtgttc	1680
aggatgataa ctgaaaacac agttgaagag aaaattgttg agagagctga aataaaactc	1740
cgcctcgata agttggctcat ccaacaaggc aggctggtag acaataaaac ggcaactcaac	1800
aaagatgaaa tgttgaatat gatccgtcac ggtgccaatc atgtatttgc cagtaaatg	1860
tctgaaatca ccgatgaaga cattgacact attttggaaa aaggcgaagc aaggacggaa	1920
gaaatgaata aaaaacttga acaactcggg gattctaatt tgaaagactt catgatggaa	1980
accccgactg agtcagttta ccaattcgaa ggagaggatt acagggaaaa gcagaaaagt	2040
ttaggaatag gaagtggat agaacctcca aaaagagaac gtaaagctaa ttacgtctgc	2100
gatgcctatt ttagggaagc attgagagta tcagaacctc aagctccaa ggcaaccgag	2160
cctcctaacc agcctatagt tcaagatttc caattcttcc ctctcgtct ctttgagcta	2220
ttggaccagg agatctatta ctccaggaaa actgtgggct acaagtttc taaaaatcct	2280
gaattaggtt ctgatgcac acgtgtccaa aaggaagaac aaagaaagat agatgaggca	2340
gaacctttat cagaagaaga actcgtgaa aaggaaaaac ttcttacgca gggttttacc	2400
aattggacta aaagagattt caaccagttt attaaagcta atgaaaaata tggctgtgat	2460
gatattgaca atatttcaaa agaagtagaa ggaaaaactc cagaagaagt aagagcttat	2520
tcagaagtgt tctgggaacg atgtaacgaa ttgcaggaca tagatcgtat catggggcag	2580
atcgacaggg gagaggctaa aattcaaagg agagcaagta ttaagaaagc tctcgataca	2640
aagatgagcc ggtacagagc ccatttcat caacttcgca tctcctacgg tacgaataag	2700
ggtaagaact ataccgagga agaagataga ttccttgtct gtatgttgca taagcttggt	2760
tttgacaagg aaaatgtgta cgaagaactt agagcgatgg tcagggtgtgc gcctcagttc	2820
agattcgact ggttcacaa atcgagaaca gccatggaat tgcagaggcg ttgtaatact	2880
ctaattactc tcacgaaag agaaaaatcag gaacttgagg agagggaaag agccgagaag	2940
aggaaaggaa gaggaagtgg gcgtggctct ggttcggta aaaggaaagg agacggttcc	3000
atttcacttc cccctcctgt ccctggccaa ggggataaga acagccccgc cagaaaaaag	3060
aaaaaaatgt ag	3072

<210> SEQ ID NO 66

<211> LENGTH: 1164

<212> TYPE: DNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 66

atgaataaaa aacttgaaca acttggtgtt gattcatcat taaaagattt catgatggag	60
gctcccactg agtctgtota tcagtttgaa ggcgaagatt atagagaaaa gcaaaaagt	120

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tttgaattg gaaattggat tgaaccacca aaacgagaac gtaaagcaaa ttatgcagta	180
gatgcctatt ttagagaagc actgagagtt tcagaaccta aagctccaaa ggcccctagg	240
ccaccaaagc aaccatagtt tcaagatttc caatttttcc cactcgtct gtttgagctg	300
ttagatcaag aaatatacta ttttcgaaaa actgtttgct acaagggttc taaaaatccg	360
gagttaggat cagatgcttc tcgtatacaa agggaagagc aaagaaaaat tgatgaagct	420
gagcgttga ctgaggaaga gctagctgag aaagaaaact tattgaccca ggggtttact	480
aattggacta aaagagattt taaccagttc ataaaagcta atgaaaaata tggacgtgat	540
gatattgata atatctcaaa agatgttgaa gggaagactc cagaagaagt acgagcatac	600
tctgaagtat tttgggaaag gtgcaatgaa ctacaggcca tagatcgat catggggcag	660
attgatagag gtgaagcgaa aattcaaaga agagccagta ttaaaaaagc ttagatata	720
aagatgagtc gatatagagc accgtttcat caactacgaa ttgcttatgg tacgaacaag	780
gggaaaaatt acacagaaga agaagacaga ttccttgtgt gcatgctaca taagcttggc	840
tttgataaag aaaatgtgta tgaggaaactt agggcgatgg tgaggtgtgc tcctcagttt	900
aggtttgatt ggttcaccaa gtctcgaaca gctttggaat tgcaaagacg ttgtaatact	960
ctaatacagt taattgaaag ggaaaaccaa gaattagaag aaagggaaaa agtagaaaaa	1020
aggaaaagtc gaggcagtaa tgggcgtggt ccagttctg gtaaacgtaa gggagatgga	1080
tctatttcat ctccacctgt ctctgtacag agtgataaaa gcagccctgc tcggaaaaag	1140
aaaaagtata tctctgttga gtaa	1164

<210> SEQ ID NO 67

<211> LENGTH: 1665

<212> TYPE: DNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 67

atgggtcttg gtaaaactat tcagactatc tgcagtttgt attatttatt tcatactcac	60
cagttgtatg gaccattttt aattgttgtc cctttatcta cgatgacttc atggcagagg	120
gagttttcat tatgggctcc agaaatgaat gttgtaactt atattggtga tataaactcc	180
cgtgatgtta tacgtaatta tgaatggtgc tattcaggtt cgaaaagggtt aaaattcaat	240
gccattctta ctacatatga aattgttctt aaagacaaag catttttggg tagtataagc	300
tgggctatcc ttatggttga tgaagcacac agattgaaaa atgatgattc attattatac	360
aaaacattga aagagtttga taccaaccat aggtcttcta taacaggcac tcctttgcaa	420
aatagtctta aagagctttg ggcgttgtct cactttatta tgcccaacag atttaataac	480
tgggaagaat ttgaaaaga acatgacaac tctgctaata aaggctatac taagttgcac	540
agacagctgg aaccatatat tctacgacga gttaagaagg atgttgagaa atctttacca	600
gctaaagtgg aacaaatatt acgtgttgaa atgacatctg tacagaagca gtattacagg	660
tggattttgt ccaaaaatta ttctgctctt cgaaaaggag tcaaagggtc tcctagtaca	720
tttataaata ttgttattga attaaaaaaa tgctgtaatc atgcacatct aataaaacca	780
ttagaaaatg aagcaaaaac tgaagactac ttacagcaat tggtaaaagg ctcagggaaa	840
ttacttctgt tggacaagtt gcttgttcgc cttaagaaa ctgggcatag agtacttata	900
ttttctcaaa tggtagcaat gttggatata ctggctgagt atcttcaaat gagacatttc	960
cctttccaac gtttagacgg ttcaattaaa ggtgaattga gaaagcaagc cctcgatcat	1020
ttcaatgctg aaaattcacc agatttctgt ttcttattat caactcgtgc tgggtggttg	1080

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ggcattaatt tagcaacagc tgatactgtc attatatattg actctgattg gaatccacaa	1140
aatgatttgc aagcacaagc tagagctcat agaatcggtc agaaaaatca ggtgaacata	1200
tacagacttg ttactaaaag ttctgttgag gaaaatattg tcgagcgggc caaacaaaaa	1260
atggtcttag atcatttagt tatacaaaga atggatacta caggtagaac tgtcctggat	1320
aaaaaaaaatt cttcatccag tgcgcctttt aacaaagaag aacttactgc tattttaaaa	1380
tttggggctg aagaattatt taaagatgaa gaagatgggtg atgaagaacc aacttgtgac	1440
attgacgaaa ttttgagaag agctgaaacc agagatgaag gaccagccac tgttggtgat	1500
gaattacttt ctgcttttaa agttgcaagt ttgcttttg atgaagataa agaaactcag	1560
agtgaaccag aacagcagga tgacgatact agagattggg tttgttatga taatacaatc	1620
ttatgtatct cgtattttat gattgaacca atttataatc aataa	1665

<210> SEQ ID NO 68

<211> LENGTH: 4569

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 68

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ccccuccac aagcucauc accuauuggc cgcgcgagg gcgccccauc gccaaugccc	120
ccuucuaacc aacaggcggc cucaccaau gguccaccgc accaccccca cagcccgaca	180
gguuaccaag gagggaucc acacaugaau ggaccaaau guguuccucc ugguaugcag	240
caggcuacuc aaacauuuc gccucaucag caauugccac cccaccagca accaccaau	300
cagacugcuc cuggugggcc ugcuauggu ggagacaag aaaauuuag cgcucuccag	360
cgugcaauag auucuaugga agagaaggc cuucagggaug auccacguua cucgcagcug	420
cuugcguuga gggcaaggca ugccaacau gaaccuccgc uuaggccucc aucucagcuu	480
guugggggug gguucagcgg ugaggguugu gcccuccuc cugcuuaaca cagcuucagc	540
gcgaaccaac ugcaacaacu ucgagugcag aucauggcgu aucgccuacu ugcuaaggaa	600
caaccucuuu cccagcagcu agcuuuggcu gugcaaggca aacgccucga cagcccggc	660
gaguccaacu accagcaucc uccuagugaa ggagcaggag guguuggugg agaaggaagu	720
ggagacgggg gaucgucgaa cggccugaug acgcagccga ugcgugcccc augcccccu	780
ggugggccagc ccccaacggc cucaccgaug acaggccaga uggcaccucc uacugggcca	840
gcuccuguaa ggccaccucc ucccgguug ucuccuacac cuccgcgcc uccucagcag	900
guuccuggug cuccgggggc cccacaacca aagcaaaa ggguuaccac caugccaaga	960
ccgcaugguu uagaucuccu ucuuuuuc caggaaagag agaauagagu agccgcuagg	1020
auuguacaua ggauggaaga auuaucaau uuaccagcua cgaugccuga agaccuucga	1080
auaaaagcgc agauaagacu uagggccuug agggguacuua acuccaaaag gcauuuaga	1140
gcagagguga uagcuuguc uagacgcgau acaacauuag aaacagcugu aaugugaaa	1200
gcuuauaaac gaacgaagag gcaaggcuua cgggaagcca gagcuacgga aaagcuugaa	1260
aaacaacaga aacuuagac agaaaaggaag aagagacaaa aacaccagga auaucugagc	1320
acuauuuugc aacauugcaa agacuucaaa gaauuccaua gaaauaauu ugcuaaaguu	1380
gguauguuaa auaggcgugu gaugaaauac caugcgaug cgcagcguga acagaagaaa	1440
gagcaagaaa ggauagaaaa agaacguau agaaaggcuua uggcugagga ugaagagggu	1500

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uacaggaac	ugauugauca	gaaaaaagau	aagagauugg	cauuccuucu	uucacaaacu	1560
gaugaaaua	uugccaaucu	uacugaaaug	gugaagcagc	auaaaugga	acaacagcgu	1620
aagcaggaac	aagaagagca	acaaaaacgg	aagaggaaaa	agaaaaagaa	gaauagggaa	1680
ggagauccag	augaugaaag	cucucagau	ucagauuuac	auguuagcgu	uauagaagca	1740
gcaacugguc	ggcagcugac	gggggaggau	gcuccauugg	ccagccagcu	ugggagcugg	1800
uuggaggcac	accgggugc	ggagccuug	gaagauagcg	aagaugaaga	ugaugaagag	1860
gacagcgacg	aggaagguga	ugauaacagu	agaucaaaag	gugguuuuuc	aaugauagga	1920
aaaugaagaag	cugauagcaa	guuauucugu	gaagacgaag	cucgagaaa	gauaaagaaa	1980
gcgaagauug	aagaugauga	auacaagaac	acgaccgaag	aacauacaua	cuacagcauc	2040
gcucacaccg	ugcaugaaau	ugucaccgaa	caagcuuca	ucaugauuaa	cgguaaaauug	2100
aaagaauauc	aaaauaaagg	ucuuagaugg	uugguuuucu	uauacaacaa	caacuugaau	2160
ggaauccugc	ccgacgagau	gggccuuggc	aagacaauuc	aaacaauagg	ucucauuacu	2220
uauuugaugg	agaagaagaa	aguaaauggu	ccuuaccuca	uuauuguucc	ucugucaaca	2280
uuauccaaau	ggguuuugga	auucgagaaa	ugggcuccuu	caguguuuugu	ggugagcuau	2340
aaagguucuc	cugcaaugag	gagaacuuua	caaucacaga	ugcgucgcag	gaaguucaau	2400
guccugcuca	cgaccuacga	guaugucauc	aaggacaagg	caguacuugc	aaaguugcau	2460
uggaaguaca	ugauaaucga	cgaggggacac	aggaugaaaa	accaccauug	uaagcugacg	2520
caggugcuga	acacccauua	uuuggcaccu	caccgccucc	uucucacggg	cacaccucuc	2580
cagaacaaac	uaccugagcu	cugggcucuu	cuaaacuuuc	uccucccguc	caucuucaag	2640
ucguguucua	cguuugagca	augguucaau	gcaccuuuug	cuaccacugg	agaaaagggu	2700
gaguugaaug	aggaagaaac	aaauuugauu	aucaggcggu	uacauaaggu	ccuucgaccu	2760
uuccuccuuc	gucgacugaa	aaaggaaguc	gaaagucagu	ugccagagaa	aauguauuac	2820
aucgucaagu	gugauauguc	uggucuccaa	cguguacuua	auaggcacau	gcagaguaaa	2880
ggaguccugc	uuaccgaugg	uucugagaag	ggcaagcagg	guaaaggagg	agcuaaagcg	2940
cuaaugaaca	cgauccgucca	auugaggaag	cuuugcauuc	auccuuucau	guuccaucau	3000
auugaagaaa	aaauuuguga	ucacguuggc	cagaacaaacg	uugucacagg	gccugaucug	3060
uuccgaguuu	cugguaaaau	ugaaauccuc	gaucguauau	ugccaaaacu	gaaggccacg	3120
agccauaggg	uacuucuuuu	cugucaaaug	acucagcuga	ugaccaucau	ggaggauuau	3180
uugucuugga	gaggguucuc	cuaccuucgu	cuugauggua	cgaccaaauc	ugaagaccga	3240
ggagauucuc	ugaaaaaaau	caacaaucca	gaaagugaau	uuuuuuuuu	cuugcucuca	3300
accagagcug	gaggucucgg	auugaacuua	caggcugcag	auacugucau	uauauuugau	3360
ucagaauugga	accucauca	ggauuuacaa	gcucaagaca	gagcucauag	gauuggacag	3420
caaaacgaag	uucguguuuu	gcggcuuaug	acaguaaaau	cuguugagga	gcguauucuu	3480
gcagcugcuc	gguaacaagcu	gaauauggau	gagaaaguca	uucaggcugg	uauuuuugac	3540
cagaaaucua	caggaaaccga	gaggcgagaa	uuucugcaaa	acaucucuca	ucaagaugau	3600
gcagaugaug	aggaauaaga	aguuccagau	gaugaaaugg	uuauucguau	gauugcgcg	3660
acagaagaug	aaaucaaccu	cuuccagaaa	aucgaauuag	aaaggaggag	ggaagaggcu	3720
aaacuuggac	cuaacaggaa	gucaaggcuu	guagaagagg	cggaauuacc	ugacuggcuu	3780
guaaagaauug	acgaugagau	ugagaagugg	acuuauagaag	aaaccgaggu	ccaaauaggga	3840
agagguaaua	ggcagaggaa	ggaaguagau	uauacagaua	guuugacuga	aaaagaauug	3900

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uuaaaggcca uugaugacaa uguaugau uuugaugacg augaagagga agagguaaaa 3960
acaaagaaaa gaggcaagag aagaagaagg ggagagggaug augaagaaga ugcaaguacu 4020
ucaaagagaa ggaaauuuc uccaucugaa acaaacuga ggaggcgauu gcguaaccuc 4080
augaacauug uuguuaagua uacugacagu gacucgagag uacucaguga accauucaug 4140
aaacuucccu cucgccauaa guaccagac uacuaugagu ugaucaagaa accuauagac 4200
aucaagagga uauuggccaa aguagaagag uguaaaauug cugacaugga ugaauuagaa 4260
aaggauuuua ugcaacuug uaaaaaugcu cagacauaca augaggaggc cucauugauc 4320
uugaagauu cgauaguauu agaaaguguu uucucuaug cucgucaaaa aguagagcag 4380
gauaaugauu cagaugauga ugaaguaaa ggugaccaag aagaugcugc aucagacacu 4440
ucauccguca aaauagaaau gaaacuaaag ccuggggagga cccgaggag ugagcuggu 4500
gguaaaagga ggagaagaaa auauaucucu gaagaugaag acgaagacca uagcgaagu 4560
uccuaaug 4569

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<210> SEQ ID NO 69

<211> LENGTH: 6222

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 69

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guuguucaac aagaaggcac ugaagaaaau ucaccugaca gugaugaaag aaguaggaua 120
gaggaaagag augacgagua ugaccugag gaugcgagga aaaaaagaa agguaaaaag 180
agaaaagcca aaggggaaag caaaaagaa aagaaacgua aaaaaggaa gaagaugau 240
agugcugaag aaagugaggg aggcggggaa gaagaaggcg auuccgauu uggaagaaaa 300
ucuaagaagu cuaaaggaa uucacaacca aaaccagugc agcaagaauu uucuggaggu 360
guaccuucag uagaagaagu uugcagccuu uuuggacuua cagauguaca gauugacuau 420
accgaaugug auuacaaaa ucugacuacg uauaaacuuu uucaacaaca uguucguccu 480
auucuuugca aggacaacca gaagguuucc aucggaaaaa ugaugaugcu cguggcugca 540
aaauggagag auuuuugcaa uuccaaucca aacgcuaac aggaaccaga uccagaagcu 600
ucagaagaac aggaauauuc uaaaccuacc aggacacgac cuucacgagu uucaacuaca 660
caaaaugaug augaagaaga cgacgaugcu gacgaacgag ggaggaaaaa gagaagugga 720
cgaaguaaaa agucaucagg aaagaagucc gcuccuccgg ccacaacca ggucccuacc 780
cucaagauga agauaggaaa aagaaaacag aaauccgaug aagaagauga agguucaguu 840
ggugccguuu cugaaaggga cucagaugcu gaaucgagc aaugcucgc agaagcugaa 900
gaaguuaaua aaccugaagg uguuguagaa gaagaagaag gugcagaggu ggcuccugua 960
ccuaagaaaa aggccaaaac gaaaauuggu aauaaaaga aaaggaaaaa gacacggacu 1020
acuaacaagu uuccagacag ugaagcuggu uaugaaacag aucaucagga cuauugugaa 1080
guuuucaac aaggagguga aaauauaua ugugauacgu gccucgagc uuaucauuug 1140
gucuguuugg aucccgaaau ggaagauacg ccagaaggca aauggucaug ccucuuugu 1200
gaaggugaag guguaacgga aaaagaagau gauguccauc aagaauuuug cagaguuuugu 1260
aaagauggug gagaacuuuu augcugugau ucuugcccuu cugcauacca cacauucugu 1320
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aagccuuuga	gagguaaagu	gucaaagauu	cuuacuugga	ggugguugga	aucucccagu	1440
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gucaaguggg	cagauauguc	uuauuggcac	uguaguuggg	ugucugaacu	ucagauggau	1560
guuuuucuaa	cucaaaugau	caggaguauu	auucguaaa	augauaugga	cgaaccuccc	1620
aaacuagaag	aaccuugga	ugaagcagac	aaugagauga	agaggauacg	agaggcaaa	1680
aucaaugagc	aagaauuaga	agagaaauau	uacaaguau	guaucaaac	agaguggcuu	1740
auugugcaga	ggguauuu	ccaucgcacu	auaagggaug	gaagcaaucu	guaccucguc	1800
aaauggaggg	accucccuu	ugaccaggcg	acuugggagg	aagaagucac	cgauaucccu	1860
ggcuugaaga	aagcuauuga	auuuuacaa	gagaugaggg	cuugcuguuu	aggugaauuc	1920
aaaaaacuaa	aaaaagguaa	agguaaaaga	ucaagagag	aucaagauga	ugagggaagg	1980
agcagaagug	caggaaugau	ggcgucggg	ggaccagcua	cuggucaaua	cuucccgccu	2040
ccugaaaagc	cugucacaga	uuugaaaaag	aaauacgaa	aacagccgga	cuaucucgac	2100
gucuccggua	ugugccuuc	uccuuaccaa	uuagaagguu	uaauugguu	gaggauuuc	2160
uggggggaag	gaacagacac	uauucuuugc	gaugagaugg	gucuuaggaa	aaccuuucag	2220
acaauuacuu	uccucuauuc	ucuuuacaaa	gagggucauu	guaaaggccc	cuuccuugug	2280
aguguacccu	uaucuacaau	uaucaauugg	gaaagagagu	ucgaaacuug	ggcgccagac	2340
uucuaacguug	ucacauaugu	cggagacaaa	gauucucgug	cuguaauacg	ugaaaugaa	2400
uuuucauucg	augauaaugc	uguuagagga	ggaagaggug	uuucuaaagu	ucgcucuucu	2460
gcaauaaagu	uucauguacu	gcuaacauuc	uaugaacuaa	ucucuaucga	ugucacuugc	2520
cuuggaucga	ucgagugggc	agugcuugua	guagaugaag	cacacaggcu	gaaaaguaau	2580
cagagcaagu	ucuuuaggcu	ucuuugcuca	uaccacauug	cuuauaaacu	ucugcugaca	2640
ggaacuccgu	ugcaaaacaa	ucuaagaaga	uuguuucauu	uacuuauuuu	ccuuacgccg	2700
gaaaaauuca	acgaccuugc	gacauuucac	aacgaauucg	cugauauuuc	aaaagaagaa	2760
caagucaaaa	gacuucauga	guuacucggg	ccgcuaugug	ugaggagauu	aaaagcugau	2820
guacucaaga	auaugccuac	aaaucucgag	uucuuuguaa	gaguugaacu	cuccccgaug	2880
cagaagaagu	acuacaaa	uauucucaca	aggauuuucg	aagcuuuaaa	uccaaaagg	2940
ggcggucaac	aaguauucuc	uuugaacauu	augauggauc	uuaaaaaau	cuguaaucau	3000
ccauaccugu	uuccugcugc	uucucaggaa	gcuccuuuag	gaccaagcgg	aucuuacgau	3060
cuucaagggu	uaaucaaacg	aucuggaaaa	ugauuacuu	ugucgaaaa	gcugagacgg	3120
cucaaaaga	aggguacac	aguacugauu	uucucucaaa	ugacaaaaau	guuggacuua	3180
uuagaagacu	accucgaggg	ugaagguuau	aaauaugaac	guauugacgg	uacgaucacc	3240
ggugacuuua	gacaagaagc	uacgaucgg	uuuacgccc	cuggagcuca	acaaauuguu	3300
uuucuuuugu	ccacucgugc	gggaggucuu	gguaauuau	ucgcucucgc	agauacaguu	3360
auuuuuuug	acucugacug	gaauccucau	aacgauuuuc	aggccuuuuc	gagagcacac	3420
aggauagggc	aagcaaacaa	gguaaugauu	uaucauuug	ugacacgagc	gucuguugaa	3480
gaaagaguua	cgcaaguggc	uaagagaaaa	augauguuua	cccaucuuug	cguacgacca	3540
gguaugggug	gcaagcaagc	aaauuucacu	aagcaagaac	uugaugauau	uuuaaggguu	3600
ggaacagaag	aacuuuucac	agaagagcag	gguaaagaag	augaagccau	ucauuuugac	3660
gauaaagcug	uugaagaauu	acuugaccgg	ucgaagaugg	guauugaaca	gaaagaaaac	3720
uggucuaaug	aaauucuuuc	uucuuuucac	guggcaaguu	auguuacuaa	agaagaagac	3780

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gaagaugagg	aaaauaggaac	agagguaaua	aaacaggaag	cagaaaauac	agaccagacu	3840
uaauuggguca	aacuguugag	gcaccauuau	gagcaacaac	aagaggauau	uucucgaacu	3900
cucgguaaag	gaaaaaggau	ucgaaaacag	gugaauuaca	ucgacggugg	agugauggac	3960
ucaagagaga	acgccgauuc	gacguggcaa	gacaaccucu	cugacuauaa	uucagacuuc	4020
ucuguccuu	cugaugauga	caaggaagac	gaugacuug	augagaaaa	ugaugaugga	4080
acgagaaaga	agcguaggcc	agaaggagg	gaggacaaag	auaggccucu	accuccucu	4140
cuugcccag	ucgguggaaa	cauugagguc	cugggauuca	acgccagaca	gcguaaagca	4200
uucuugaaug	cuauuugag	guauggaug	ccaccucaag	augcauucaa	cucgcagugg	4260
cuuguucgag	accugagggg	uaauucugag	aagcauuuca	aggcauacgu	auccucu	4320
augaggcauu	ugugugagcc	uggcgaggac	aaugccgaaa	cauucgagg	ugguguucca	4380
agggagagguc	uuagucggca	gcauguucuc	acaaggauag	gugugauguc	acucauuagg	4440
aaaaagguuc	aagaauuuga	gcaauuuau	ggauuuacu	cgaugccuga	aauguugaag	4500
aaaccacuug	uugaugccgg	auugcauaaa	acaagugcua	gcaguauagg	ugaaggugcu	4560
aguaguuccg	guacaccugc	aacaucagcu	gcuccaaguc	cagcuccuac	ucuuuuggau	4620
aagacacaaa	uugaagauuu	gagugaaaaa	gaagauccgu	caaagacuga	agauaaaacc	4680
accgaugauu	ccaaaccuc	agaagaggcu	aaagcugcag	augaugcaaa	uaagccucag	4740
gcugaaggag	aaaaggcaga	aggauucucu	aaugcaaac	aaacuucuga	agcugaagga	4800
agcgaugaga	aaaaacccaa	agaagaaccg	auggauguag	auggugaagg	agaggcuaaa	4860
gauaugugaua	agacagaaaa	acaagaaggu	acugacgaaa	aagauguagc	ccuaaaagag	4920
gaagaaaagg	augaagaggu	caacaaagag	aaggagagg	aaacagagga	aaagaagguu	4980
aucgaauuuug	aagaagacaa	aucuaaaagg	aaauuuauug	ucaauaucgc	ugauggagga	5040
uuuacugagc	uccauaccuu	auggcaaaau	gaagagaaag	cugcaguacc	ugguagggag	5100
uacgagaucu	ggcauaggag	gcaugacuau	uggcuguugg	guggaaucgu	uacccauggc	5160
uauggucggg	ggcaagauau	ucaaaaugau	auuagauuuug	cuauuuacaa	cgaaccauuu	5220
aagauggaug	uuggaaaagg	aaauuucua	gaaauuaaaa	auaaauuuc	ugccaggagg	5280
uuuaagcuuc	uugagcaagc	ucuggugauu	gaagaacagu	uaagacgugc	agcuuuuuu	5340
aaucugacgc	aagauccaaa	ucaccagca	augucacuga	augcaagauu	ugcagagguu	5400
gaaugucuag	ccgaauuca	ccaaccacc	ucgaaggaaa	gucucgugg	caacaaaccu	5460
gcaaaugcag	uguuacauaa	aguauugaac	cauuuagagg	agcuucuguc	ggauaugaaa	5520
ucugacguau	cucgacuacc	agccacucua	gccagaauuc	caccuguagc	ccagaggcua	5580
cagaugucug	aacggucaau	acuucuaagg	uuggcugcaa	cuacuucucc	ugcgacgcc	5640
accacgucgc	aucaaacugg	uaugauaagc	agucaguucc	cugcuggaau	ucaaucaggg	5700
caguugacug	gaacguuucc	gaaugccagu	uuuaccaacu	ucaggcccca	guauucaguu	5760
ccugggcaaa	cugcagccca	ggguuuucc	gguaauugau	aaugaaagc	uggacgguaa	5820
uugucugcga	gugaauucuc	caugaguaaa	uaauaggguu	uuuuuuuuu	uuagaaaga	5880
aaauaaagaa	gcguuuuugu	uaguuuuguu	gauaguucuc	uuuuuuucuu	ucauuuuugu	5940
uuuagcggaa	aaaaaaugu	ucauuuaag	uaacuuaaa	auuggacaug	cuauuuuuu	6000
uuccuuuag	auuuuuugu	uuuuuguuag	uuuuucggua	uuguaagaa	gucuauuugu	6060
guaagagguu	guacaagauu	gccuaaauc	cuuguauuu	uuuuuuuac	uuuugaaua	6120

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aaaaaaaaa uauuuuacuu cgaucuuagg uuaaggguaa uaaaaaaaaa uguuacugga 6180
aaaaaaaaua gaaaaaaaua aaaagauagc cuuuccccuu ac 6222

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<210> SEQ ID NO 70
<211> LENGTH: 3072
<212> TYPE: RNA
<213> ORGANISM: Euchistus heros

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<400> SEQUENCE: 70

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augucgaagc caaaugaagu uaguuuuggau acaacagaua cuguugaaa uucuaaugaa 60
ucuucgggag acacagaguc guccaagggu aaaaaugaag auuuugaaac aaaaauugaa 120
acugaccguu cuagaagauu ugaguucug uugaagcaga cagaaauuuu uucacauuuu 180
augacaauc aaggaaaguc gaacagcccu gcaaagccua aagucggccg uccuagaaag 240
gaaacuaaua auuuggcacc agccgguggu gaugguucug ccgaccaucg gcaucguaug 300
accgagcagg aagaagauga agaacugcuu gcugaaagua auacuucuc aaaauccuaa 360
gcaagguuug acgcuucucc uuuuuauuu aaaagcggag aguugaggga uuaccagaua 420
cgugguuuga auuggaugau aucccucua acacacgguu uaaugguau acuugcugau 480
gagauggguu uagguaaaac ucuccaaacu auuucucucc uggguuacau gaagcauuau 540
agaaauauac cagggccaca uauggucauc guacccaaau caacauuagc uaauggaug 600
aauaauuuu aaaaguggug cccaaccucg cgugcugucu guuuauucgg agaucaggaa 660
acgagggaug cguucaucag agacacucuu augccgggug aaugggaugu cugcguuaca 720
ucuuaugaaa ugaucaucg agaaaagagc guuuucaaga aguucaacug gagguauaug 780
gucauugacg aagcccacag gaucaagaau gaaaaaucca aacucuccga gauugugaga 840
gaguucaaaa cgacgaucg auuacuccug accgguacuc cuuuacaaa uaccuccac 900
gaauguggu cucuucuaa cuuccucua ccagauguu ucaaucauc agaugauuu 960
gauucauggu uuaauacca uaccuuccu ggcgaauuu cucuugucga gagauuacau 1020
gcugucuga gaccuuuccu ccuaagaaga uugaaaucug agguagagaa aaaacucaaa 1080
ccgaagaaag aagucaaaau cuacguugga uugaguaaaa ugcagagaga augguauacu 1140
aaaguucuaa ugaagaauu agacauugua aacggugcug gccgagucga aaaaugcgc 1200
cuccaaaaca uccucaugca guugagggaag ugcaguauc acccuuacu cuucgacgga 1260
gcugaaccag guccaccuaa cucaacugau gagcaucug uauauaacag uggaaaaug 1320
guaauuuag acaagcuuc uccuaauug caagaacaag gaucacgagu ucugguuuu 1380
agccaaauga caaggauu ugaauuuc gaagauuacu guuauuggag aggauauau 1440
uacugucguc ugaugguua uacaccucau gaggauaggc agagacagau uaaugaguuc 1500
aacgaagaag acaguaagaa auucauuuuc auguugucga cucgugcggg ugguuugggu 1560
aucauuuag ccaccgcaga uguagucau uuguacgau cggauuggaa cccucaaaug 1620
gaucuccagg cuauggaucg ugcucaucgu auuggucaaa agaaacaagu caaaguguuc 1680
aggauauaa cugaaaacac aguugaagag aaaaauugu agagagcuga aauaaaacuc 1740
cgccucgaua aguuggucau ccaacaaggc aggcugguag acauuuuuac ggcacucaac 1800
aaagaugaaa uguugaauu gaucgucac ggugccauc auguauuugc caguuuagau 1860
ucugaaauca ccgaugaaga cauugacacu auuuuggaaa aaggcgaagc aaggacggaa 1920
gaaugaaua aaaaacuuga acaacucggu gauucuaau ugaagacuu caugauggaa 1980
accccgacug agucaguuaa ccaauucgaa ggagaggauu acagggaaaa gcagaauguu 2040

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uuaggaauag gaaguuggau agaaccucca aaaagagaac guaaagcuaa uuacgcuguc 2100
gaugccuauu uuagggaaagc auugagagua ucagaaccua aagcucccaa ggcaccgagg 2160
ccuccuaaac agccuauagu ucaagauuuc caauucuuc cuccucgucu cuuugagcua 2220
uuggaccagg agaucuaaua cuucaggaaa acugugggcu acaaaguucc uaaaaauccu 2280
gaauuagguu cugaugcauc acguguccaa aaggaagaac aaagaaagau agaugaggca 2340
gaaccuuuau cagaagaaga acucgcugaa aaggaaaaac uucuuacgca ggguuuuacc 2400
aaauggacua aaagagaauu caaccaguuu auuaaagcua augaaaaua uggucgugau 2460
gauauugaca auauuucaaa agaaguagaa ggaaaaacuc cagaagaagu aagagcuau 2520
ucagaagugu ucugggaaagc auguaacgaa ugcaggaca uagaucguau cauggggcag 2580
aucgacaggg gagaggcuua aauucaagg agagcaagua uuaagaaagc ucucgauaca 2640
aagaugagcc gguacagagc ccuauuucac caacuucgca ucuccuacgg uacgaauaag 2700
gguaagaacu auaccgagga agaagauaga uuccuugucu guauguugca uaagcuuggu 2760
uuugacaagg aaaaugugua cgaagaacuu agagcgauug ucaggugugc gccucaguuc 2820
agaaucgacu gguucaucaa aucgagaaca gccauggaau ugcagaggcg uuguauuacu 2880
cuauuacuc ucaucgaag agaaaaucag gaacuugagg agagggaaag agccgagaag 2940
aggaaaggaa gaggaagugg gcgugguccu gguuccgguu aaaggaaagg agacgguucc 3000
auuucacuc cccuccugu cccuggccaa ggggauaaga acagccccgc cagaaaaaag 3060
aaaaaaugu ag 3072

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<210> SEQ ID NO 71

<211> LENGTH: 1164

<212> TYPE: RNA

<213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 71

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augaauaaaa aacuugaaca acuugguguu gauucaucau uaaagauuu caugauggag 60
gcucccacug agucugucua ucaguugaa ggcgaagau auagagaaa gcaaaaugu 120
uuuggaauug gaaauuggau ugaaccacca aaacgagaac guaaagcaa uuaugcagua 180
gaugccuauu uuagagaagc acugagagu ucagaaccua aagcuccaaa ggcccuagg 240
ccaccaaagc aaccuauagu ucaagauuuc caauuuuucc caccucgucu guuugagcug 300
uuagaucag aauauuacua uuucgaaaa acuguuugcu acaagguucc uaaaaauccg 360
gaguauagau cagaugcuuc ucguauacaa agggagagc aaagaaaau ugaugaagcu 420
gagccguuga cugaggaaga gcuagcugag aaagaaaacu uauugacca ggguuuuacu 480
aaauggacua aaagagaauu uaaccaguuc auaaaagcua augaaaaua uggacgugau 540
gauauugaua auaucucaa agauuugaa gggaagacuc cagaagaagu acgagcauac 600
ucugaaguau uuugggaaag gugcaaugaa cuacaggcca uagaucguau cauggggcag 660
auugauagag gugaagcgaa aauucaaga agagccagua uuaaaaaagc uuugauaca 720
aagaugaguc gauauagagc accguuucac caacuacgaa uugcuuauug uacgaacaag 780
gggaaaaauu acacagaaga agaagacaga uuccuugugu gcaugcuaca uaagcuuggc 840
uuugauaaag aaaaugugua ugaggaacuu agggcgauug ugaggugugc uccucaguuu 900
agguuugauu gguucaucaa gucucgaaca gcuuugaaug ugcgaagacg uuguauuacu 960
cuauucacgu uaaaugaag ggaaaaccaa gaauugaag aaagggaaga aguagaaaa 1020

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aggaaaaguc gaggcaguaa ugggcguggu cccaguucug guaaacguua gggagaugga	1080
ucuauuucau cuccaccugu cucuguacag agugauaaaa gcagcccugc ucggaaaaag	1140
aaaaaguaua ucucuguuga guaa	1164

<210> SEQ ID NO 72
 <211> LENGTH: 1665
 <212> TYPE: RNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 72

auggggucuu guaaaacuau ucagacuau ucaguuugu auuuuuuuu ucauacucac	60
caguuguau gaccuuuuu auuuguuguc cuuuuauca cgaugacuuc auggcagagg	120
gaguuuucau uaugggcucc agaaaugaau guuguacu uauuuguga uauaaacucc	180
cgugauguua uacguauua ugaauugugc uauucagguu cgaaaagguu aaaaaucau	240
gccauucua cuacauaaga auuuguucuu aaagacaaag cauuuuuggg uaguauaagc	300
ugggcuaucc uuaugguuga ugaagcacac agauugaaaa augaugauuc auuuuuuac	360
aaaacauuga aagaguuga uaccaaccu aggcuuucua uaacaggcac uccuuugcaa	420
aaugucuaa aagagcuug ggcguugcuu cacuuuuu ugcaccaacg auuuuuuac	480
ugggaagaau uugaaaaaga acaugacaac ucugcuaua aaggcuauac uaaguugcac	540
agacagcugg aaccuuuuu ucuacgacga guuaagaagg auguugagaa aucuuuacca	600
gcuaaagugg aacaaauuu acguguugaa augacaucug uacagaagca guuuuacagg	660
uggauuuugu ccaaaaauu uucugcucu cgaaaaggag ucaaagguu uccuaguaca	720
uuuuuuuuu uuguuuuuga auuuuuuuu ugcuguauc augcacauc auuuuuuacc	780
uuagaaaau aagcaaaac ugaagacuac uuacagcau uguuuuagg cucagggaaa	840
uuacuucugu uggacaagu gcuuguucgc cuuuuagaaa cugggcuaug aguacuuua	900
uuuuuucuaa ugguacgaau guuggaaua cuggcugagu aucuuucaa gagacuuuc	960
ccuuuccaac guuuagacgg uucauuuuu ggugaauuga gaaagcaagc ccucgauc	1020
uucaaugcug aaaaauacc agauuuucgu uucuuuuu caacucgugc uggugguuug	1080
ggcauuuuu uagcaacagc ugaucuguc auuuuuuuu acucugauug gaauccaca	1140
aaugauuugc aagcacaagc uagagcuau agaauagguc agaaaauca ggugaacau	1200
uacagacuug uuacuuuag uucuguugag gaaaauuug ucgagcgggc caaacaaaa	1260
auggcuuag aucauuuagu uauacaaaga auggaucua cagguagaac uguccugau	1320
aaaaaaaaa cuuauccag ugcgcuuuu aacaaagaag aacuuacugc uuuuuuuuu	1380
uuuggggcug aagaauuuu uaaagauga gaagauggug augaagaacc aacuugugac	1440
auugacgaaa uuugagaag agcugaaacc agagaugaag gaccagccac uguugguau	1500
gaauuacuuu cugcuuuua aguugcaagu uuugcuuuu augaagaua agaaucucag	1560
agugaaccag aacagcagga ugacgauacu agagauggg uuuguuuga uauuacau	1620
uuauuauuc cguuuuuu gauugaacca auuuuuuac auuaa	1665

<210> SEQ ID NO 73
 <211> LENGTH: 1134
 <212> TYPE: DNA
 <213> ORGANISM: *Euchistus heros*

<400> SEQUENCE: 73

tacaaaatgt gtgacgaaga agttgctgct ttagttgtag acaatggatc tggtatgtgc	60
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aaagccggtt tcgctggaga tgatgcaccc cgagctgtat tcccatcaat tgttggcagg	120
cctagacacc aggggtgtcat ggttggaaatg ggacaaaagg acagtatatgt tggagacgaa	180
gcccaaagca agagagggtat cctcaccctg aaatacccca ttgaacacgg tatcatcacc	240
aactgggacg acatggaaaa gatctggcat cacaccttct acaacgagct gcgagtcgct	300
ccagaggaac accccatcct cctgactgag gctccctca accccaaagc caacaggag	360
aagatgaccc agatcatggt tgagaccttc aacacccag ccatgtatgt cgccatccag	420
gctgtactct ccctctatgc ctccggctgt actaccgta ttgtactga ctccaggagat	480
ggtgtctccc acaccgtacc catctatgaa ggttatgccc tccccacgc catectcgt	540
ctggatcttg ctggacgtga ctgactgac tatcttatga agatcctcac cgagcgtggt	600
tacagcttca ccaccacgc tgaaggga atcgtcagg acatcaagga aaaactgtgc	660
tatgtcgccc tggactttga gcaggaaatg gccaccgccc ctgctccac ctccctggag	720
aagtcctatg aacttccga cggtcaggtc atcaccatcg gtaacgagag gttccgttgc	780
ccagaggctc tcttcacgc ttcctcttg ggtatggaat cttgcggtat ccatgagact	840
gtctacaact ccatcatgaa gtgcgacgtt gacatcagga aggacttgta cgccaacacc	900
gtcctctccg gaggtaccac catgtacca ggtattgctg acaggatgca gaaggaaatc	960
accgccctcg ctcttcaac catcaagatc aagatcattg ctccccaga aaggaagtac	1020
tccgtatgga tcggtgggtc catcttggt tccctgtcca ccttcagca gatgtggatc	1080
tccaagcagg aatacgacga atccggccca ggcacgtcc accgcaaattg cttc	1134

<210> SEQ ID NO 74
 <211> LENGTH: 22
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Primer Actin42A-F

<400> SEQUENCE: 74

tcaaggaaaa actgtgctat gt	22
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<210> SEQ ID NO 75
 <211> LENGTH: 20
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Primer Actin42A-R

<400> SEQUENCE: 75

taccgatggt gatgacctga	20
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<210> SEQ ID NO 76
 <211> LENGTH: 12
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: Probe Actin42A-FAM

<400> SEQUENCE: 76

accgccgctg cc	12
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<210> SEQ ID NO 77
 <211> LENGTH: 19
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:

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<223> OTHER INFORMATION: Probe brm-F

<400> SEQUENCE: 77

tcatcaagga caaggcagt

19

<210> SEQ ID NO 78

<211> LENGTH: 21

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer brm-R

<400> SEQUENCE: 78

gacgggagga gaaagtttag a

21

<210> SEQ ID NO 79

<211> LENGTH: 19

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Probe brm-FAM

<400> SEQUENCE: 79

cgacgaggga cacaggatg

19

<210> SEQ ID NO 80

<211> LENGTH: 17

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer mi-2-F

<400> SEQUENCE: 80

gatgagggt tgctgtt

17

<210> SEQ ID NO 81

<211> LENGTH: 18

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer mi-2-R

<400> SEQUENCE: 81

gaggcgggaa gtattgac

18

<210> SEQ ID NO 82

<211> LENGTH: 22

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Probe mi-2-FAM

<400> SEQUENCE: 82

atgaggaagg aagcagaagt gc

22

<210> SEQ ID NO 83

<211> LENGTH: 23

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Primer iswi-1-F

<400> SEQUENCE: 83

gagttcaacg aagaagacag taa

23

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<210> SEQ ID NO 84
<211> LENGTH: 19
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer iswi-R

<400> SEQUENCE: 84

cgatgagcac gatccatag 19

<210> SEQ ID NO 85
<211> LENGTH: 22
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Probe iswi-1-FAM

<400> SEQUENCE: 85

ttagccaccg cagatgtagt ca 22

<210> SEQ ID NO 86
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer iswi-2-F_MGB

<400> SEQUENCE: 86

acgtaaggga gatggatcta ttcca 25

<210> SEQ ID NO 87
<211> LENGTH: 23
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer iswi-2-R_MGB

<400> SEQUENCE: 87

cagggctgct tttatcactc tgt 23

<210> SEQ ID NO 88
<211> LENGTH: 15
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Probe iswi-2-FAM_MGB

<400> SEQUENCE: 88

ctccacctgt ctctg 15

<210> SEQ ID NO 89
<211> LENGTH: 20
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer chd1-F

<400> SEQUENCE: 89

caacagtggc tggctcttca 20

<210> SEQ ID NO 90
<211> LENGTH: 23
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Primer chd1-R

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<400> SEQUENCE: 90

accaacttgt gacattgacg aaa

23

<210> SEQ ID NO 91

<211> LENGTH: 16

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: Probe chd1-FAM

<400> SEQUENCE: 91

tctggtttca gctctt

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What may be claimed is:

1. An isolated nucleic acid molecule comprising a polynucleotide operably linked to a heterologous promoter, wherein the polynucleotide encode a double-stranded ribonucleic acid (dsRNA) molecule, the polynucleotide comprising:

a first nucleotide sequence selected from the group consisting of: SEQ ID NO:1; SEQ ID NO:63; the complement of SEQ ID NO:1; the complement of SEQ ID NO:63; at least 26 contiguous nucleotides of SEQ ID NO:1; the complement of at least 26 contiguous nucleotides of SEQ ID NO:1; at least 26 contiguous nucleotides of SEQ ID NO:63; the complement of at least 26 contiguous nucleotides of SEQ ID NO:63; at least 26 contiguous nucleotides of a native coding sequence of *Euschistus heros* comprising SEQ ID NO:3; the complement of at least 26 contiguous nucleotides of a native coding sequence of *Euschistus heros* comprising SEQ ID NO:3; at least 23 contiguous nucleotides of SEQ ID NO:3; and the complement of at least 23 contiguous nucleotides of SEQ ID NO:3;

a second nucleotide sequence; and

a third nucleotide sequence that is the reverse complement of the first nucleotide sequence, wherein the third nucleotide sequence is linked to the first nucleotide sequence by the second nucleotide sequence.

2. The nucleic acid molecule of claim 1, wherein the heterologous promoter is functional in a plant cell, and wherein the molecule is a plant transformation vector.

3. The nucleic acid molecule of claim 1, wherein the polynucleotide comprises at least 23 contiguous nucleotides of SEQ ID NO:3 or the complement of at least 23 contiguous nucleotides of SEQ ID NO:3.

4. The nucleic acid molecule of claim 1, wherein the heterologous promoter is functional in a plant cell.

5. A double-stranded ribonucleic acid (dsRNA) molecule comprising a first, a second, and a third ribonucleotide sequence,

wherein the first ribonucleotide sequence is SEQ ID NO:43; SEQ ID NO:68; the complement of SEQ ID NO:43; the complement of SEQ ID NO:68; at least 26 contiguous nucleotides of SEQ ID NO:43; the complement of at least 26 contiguous nucleotides of SEQ ID NO:43; at least 26 contiguous nucleotides of SEQ ID NO:68; the complement of at least 26 contiguous nucleotides of SEQ ID NO:68; at least 26 contiguous nucleotides of a native messenger RNA (mRNA) of *Euschistus heros* comprising SEQ ID NO:44; the complement of at least 26 contiguous nucleotides of a

native mRNA of *Euschistus heros* comprising SEQ ID NO:44; at least 23 contiguous nucleotides of SEQ ID NO:44; and complement of at least 23 contiguous nucleotides of SEQ ID NO:44,

wherein the third ribonucleotide sequence is linked to the first ribonucleotide sequence by the second ribonucleotide sequence, and

wherein the third ribonucleotide sequence is substantially the reverse complement of the first ribonucleotide sequence, such that the first and the third ribonucleotide sequences hybridize in the stem of a hairpin structure in the molecule.

6. The dsRNA molecule of claim 5, wherein the first ribonucleotide sequence comprises at least 26 contiguous nucleotides of a ribonucleotide sequence selected from the group consisting of SEQ ID NO:43, the complement of SEQ ID NO:43, SEQ ID NO:68, and the complement of SEQ ID NO:68.

7. A prokaryotic cell comprising the nucleic acid molecule of claim 1.

8. A eukaryotic cell comprising the nucleic acid molecule of claim 1.

9. A plant cell comprising the nucleic acid molecule of claim 4.

10. A transgenic plant comprising the plant cell of claim 9.

11. A seed of the plant of claim 10, wherein the seed comprises the polynucleotide.

12. A commodity product produced from the plant of claim 10, wherein the commodity product comprises a detectable amount of the polynucleotide.

13. The plant cell of claim 9, wherein the cell is a *Zea mays* cell, a *Glycine max* cell, or a cell from a *Gossypium* sp.

14. The transgenic plant of claim 10, wherein the plant is maize, soybean, or cotton.

15. A method for controlling a *Euschistus heros* insect pest population, the method comprising feeding an insect of the population with an agent comprising the dsRNA molecule of claim 5.

16. The method according to claim 15, wherein the insect pest is a male *Euschistus heros* insect pest.

17. The method according to claim 15, wherein the insect pest is a female *Euschistus heros* insect pest, the method further comprising releasing the female *Euschistus heros* insect pest into the pest population.

18. The method according to claim 15, wherein the agent is a sprayable formulation.

19. The method according to claim 15, wherein the agent is a *Euschistus heros* host plant or a part thereof, comprising a polynucleotide that is expressed in the plant to produce the dsRNA molecule.

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20. A method for improving the yield of a corn, soybean, or cotton crop, the method comprising:

cultivating in the crop the transgenic plant of claim 10.

21. The method according to claim 20, wherein the transgenic plant is a corn, soybean, or cotton plant.

22. A method for producing a transgenic plant cell, the method comprising:

transforming a plant cell with the nucleic acid molecule of claim 4;

culturing the transformed plant cell under conditions sufficient to allow for development of a plant cell culture comprising a plurality of plant cells comprising the polynucleotide; and

selecting a transgenic plant cell that has integrated the polynucleotide into its genome, and that expresses the dsRNA molecule.

23. A method for producing a transgenic plant, the method comprising:

regenerating a transgenic plant from the plant cell of claim 9.

24. The nucleic acid molecule of claim 1, further comprising a polynucleotide encoding an insecticidal polypeptide from *Bacillus thuringiensis*, *Alcaligenes* spp., or *Pseudomonas* spp.

25. The nucleic acid molecule of claim 24, wherein the insecticidal polypeptide is from *B. thuringiensis*, and is selected from a group comprising Cry1A, Cry2A, Cry3A, Cry11A, and Cry51A.

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26. The plant cell of claim 9, wherein the cell comprises a polynucleotide encoding an insecticidal polypeptide from *Bacillus thuringiensis*, *Alcaligenes* spp., or *Pseudomonas* spp.

27. The plant cell of claim 26, wherein the insecticidal polypeptide is from *B. thuringiensis*, and is selected from a group comprising Cry1A, Cry2A, Cry3A, Cry11A, and Cry51A.

28. The transgenic plant of claim 10, wherein the plant comprises a polynucleotide encoding an insecticidal polypeptide from *Bacillus thuringiensis*, *Alcaligenes* spp., or *Pseudomonas* spp.

29. The transgenic plant of claim 28, wherein the insecticidal polypeptide is from *B. thuringiensis*, and is selected from a group comprising Cry1A, Cry2A, Cry3A, Cry11A, and Cry51A.

30. The method according to claim 15, wherein the method further comprises feeding the insect of the population with an insecticidal polypeptide from *Bacillus thuringiensis*, *Alcaligenes* spp., or *Pseudomonas* spp.

31. The method according to claim 30, wherein the insecticidal polypeptide is from *B. thuringiensis*, and is selected from a group comprising Cry1A, Cry2A, Cry3A, Cry11A, and Cry51A.

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